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Search Results - Record(s) 1 through 10 of 47 returned.

1. Document ID: US 6472516 B1

L2: Entry 1 of 47

File: USPT

Oct 29, 2002

US-PAT-NO: 6472516

DOCUMENT-IDENTIFIER: US 6472516 B1

TITLE: Progestin-regulated gene

| | | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|--------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KMPC | Drawn Desc |
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2. Document ID: US 6451603 B1

L2: Entry 2 of 47

File: USPT

Sep 17, 2002

US-PAT-NO: 6451603

DOCUMENT-IDENTIFIER: US 6451603 B1

TITLE: Ribozyme nucleic acids and methods of use thereof for controlling viral pathogens

| | | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|--------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KMPC | Drawn Desc |
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3. Document ID: US 6407314 B1

L2: Entry 3 of 47

File: USPT

Jun 18, 2002

US-PAT-NO: 6407314

DOCUMENT-IDENTIFIER: US 6407314 B1

TITLE: Microspore-specific promoter from tobacco

| | | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|--------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KMPC | Drawn Desc |
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4. Document ID: US 6365159 B1

L2: Entry 4 of 47

File: USPT

Apr 2, 2002

US-PAT-NO: 6365159

DOCUMENT-IDENTIFIER: US 6365159 B1

TITLE: Spo-rel

| | | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|--------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KMC | Draw Desc |
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5. Document ID: US 6312903 B1

L2: Entry 5 of 47

File: USPT

Nov 6, 2001

US-PAT-NO: 6312903

DOCUMENT-IDENTIFIER: US 6312903 B1

TITLE: Simulataneous detection, identification and differentiation of eubacterial taxa using a hybridization assay

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

6. Document ID: US 6306657 B1

L2: Entry 6 of 47

File: USPT

Oct 23, 2001

US-PAT-NO: 6306657

DOCUMENT-IDENTIFIER: US 6306657 B1

TITLE: Polynucleotide probe and kit for amplifying signal detection in hybridization assays

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

7. Document ID: US 6299880 B1

L2: Entry 7 of 47

File: USPT

Oct 9, 2001

US-PAT-NO: 6299880

DOCUMENT-IDENTIFIER: US 6299880 B1

TITLE: Cell surface protein compounds

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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8. Document ID: US 6291204 B1

L2: Entry 8 of 47

File: USPT

Sep 18, 2001

US-PAT-NO: 6291204

DOCUMENT-IDENTIFIER: US 6291204 B1

** See image for Certificate of Correction **

TITLE: Fermentative carotenoid production

| | | | | | | | | | |
|-----------------------|-----------------------|--------------------------|-----------------------|------------------------|--------------------------------|----------------------|---------------------------|---------------------------|-----------------------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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9. Document ID: US 6277577 B1

L2: Entry 9 of 47

File: USPT

Aug 21, 2001

US-PAT-NO: 6277577

DOCUMENT-IDENTIFIER: US 6277577 B1

**** See image for Certificate of Correction ****

TITLE: Hybridization probes derived from the spacer region between the 16s and 23s RRNA genes for the detection of non-viral microorganisms

| | | | | | | | | | |
|-----------------------|-----------------------|--------------------------|-----------------------|------------------------|--------------------------------|----------------------|---------------------------|---------------------------|-----------------------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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10. Document ID: US 6271440 B1

L2: Entry 10 of 47

File: USPT

Aug 7, 2001

US-PAT-NO: 6271440

DOCUMENT-IDENTIFIER: US 6271440 B1

**** See image for Certificate of Correction ****

TITLE: Plant regulatory proteins III

| | | | | | | | | | |
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| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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| PROBE.USPT. | 111239 |
| PROBES.USPT. | 59010 |
| RRNA.USPT. | 2903 |
| RRNAS.USPT. | 307 |
| (1 AND (RRNA SAME PROBE)).USPT. | 47 |
| (L1 AND (PROBE SAME RRNA)).USPT. | 47 |

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Search Results - Record(s) 11 through 20 of 47 returned.

11. Document ID: US 6221582 B1

L2: Entry 11 of 47

File: USPT

Apr 24, 2001

US-PAT-NO: 6221582

DOCUMENT-IDENTIFIER: US 6221582 B1

TITLE: Polynucleic acid sequences for use in the detection and differentiation of prokaryotic organisms

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
| Image | | | | | | | | | |

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12. Document ID: US 6169169 B1

L2: Entry 12 of 47

File: USPT

Jan 2, 2001

US-PAT-NO: 6169169

DOCUMENT-IDENTIFIER: US 6169169 B1

TITLE: PNA probes for detection of *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
| Image | | | | | | | | | |

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13. Document ID: US 6153433 A

L2: Entry 13 of 47

File: USPT

Nov 28, 2000

US-PAT-NO: 6153433

DOCUMENT-IDENTIFIER: US 6153433 A

TITLE: Inhibitor for viral replication

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
| Image | | | | | | | | | |

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14. Document ID: US 6146855 A

L2: Entry 14 of 47

File: USPT

Nov 14, 2000

US-PAT-NO: 6146855

DOCUMENT-IDENTIFIER: US 6146855 A

TITLE: Method for the detection of viable Cryptosporidium parvum oocysts

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
| Image | | | | | | | | | | | |

15. Document ID: US 6110681 A

L2: Entry 15 of 47

File: USPT

Aug 29, 2000

US-PAT-NO: 6110681

DOCUMENT-IDENTIFIER: US 6110681 A

TITLE: Primers and probes for the amplification, detection and typing of *Mycoplasma pneumoniae*

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
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16. Document ID: US 6090551 A

L2: Entry 16 of 47

File: USPT

Jul 18, 2000

US-PAT-NO: 6090551

DOCUMENT-IDENTIFIER: US 6090551 A

TITLE: Detection of bacteria of genus *Listeria* using nucleic probe hybridization techniques

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
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17. Document ID: US 6063603 A

L2: Entry 17 of 47

File: USPT

May 16, 2000

US-PAT-NO: 6063603

DOCUMENT-IDENTIFIER: US 6063603 A

**** See image for Certificate of Correction ****

TITLE: Nucleic acid amplification process

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
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18. Document ID: US 6046006 A

L2: Entry 18 of 47

File: USPT

Apr 4, 2000

US-PAT-NO: 6046006

DOCUMENT-IDENTIFIER: US 6046006 A

TITLE: Sequential hybridization of fungal cell DNA and method for the detection of

fungal cells in clinical material

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

19. Document ID: US 6037122 A

L2: Entry 19 of 47

File: USPT

Mar 14, 2000

US-PAT-NO: 6037122

DOCUMENT-IDENTIFIER: US 6037122 A

TITLE: Nucleotide fragment of the 16S ribosomal RNA of corynebacteria, derived probes and primers, reagent and method of detection

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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20. Document ID: US 6025132 A

L2: Entry 20 of 47

File: USPT

Feb 15, 2000

US-PAT-NO: 6025132

DOCUMENT-IDENTIFIER: US 6025132 A

TITLE: Probes targeted to rRNA spacer regions, methods and kits for using said probes, for the detection of respiratory tract pathogens

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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| RRNA.USPT. | 2903 |
| RRNAS.USPT. | 307 |
| (1 AND (RRNA SAME PROBE)).USPT. | 47 |
| (L1 AND (PROBE SAME RRNA)).USPT. | 47 |

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Search Results - Record(s) 21 through 30 of 47 returned.

21. Document ID: US 5985563 A

L2: Entry 21 of 47

File: USPT

Nov 16, 1999

US-PAT-NO: 5985563

DOCUMENT-IDENTIFIER: US 5985563 A

TITLE: Detection of Ribosomal RNA using PNA probes

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Drawn Desc |
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22. Document ID: US 5976791 A

L2: Entry 22 of 47

File: USPT

Nov 2, 1999

US-PAT-NO: 5976791

DOCUMENT-IDENTIFIER: US 5976791 A

TITLE: Nucleotide fragments capable of hybridizing specifically to rickettsia rDNA or rRNA and their use as probes or primers

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Drawn Desc |
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23. Document ID: US 5955267 A

L2: Entry 23 of 47

File: USPT

Sep 21, 1999

US-PAT-NO: 5955267

DOCUMENT-IDENTIFIER: US 5955267 A

** See image for Certificate of Correction **

TITLE: Probe and method for detecting yeast of species Candida krusei

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Drawn Desc |
| Image | | | | | | | | | | | |

24. Document ID: US 5945282 A

L2: Entry 24 of 47

File: USPT

Aug 31, 1999

US-PAT-NO: 5945282

DOCUMENT-IDENTIFIER: US 5945282 A

** See image for Certificate of Correction **

TITLE: Hybridization probes derived from the spacer region between the 16S and 23S rRNA genes for the detection of non-viral microorganisms

| | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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25. Document ID: US 5853998 A

L2: Entry 25 of 47

File: USPT

Dec 29, 1998

US-PAT-NO: 5853998

DOCUMENT-IDENTIFIER: US 5853998 A

TITLE: Probe for diagnosing Enterococcus faecalis

| | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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KMC Draw Desc

26. Document ID: US 5827661 A

L2: Entry 26 of 47

File: USPT

Oct 27, 1998

US-PAT-NO: 5827661

DOCUMENT-IDENTIFIER: US 5827661 A

TITLE: Enhancing detection polymerase chain reaction assays by RNA transcription and immunodetection of RNA:DNA hybrids

| | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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KMC Draw Desc

27. Document ID: US 5801234 A

L2: Entry 27 of 47

File: USPT

Sep 1, 1998

US-PAT-NO: 5801234

DOCUMENT-IDENTIFIER: US 5801234 A

TITLE: Polynucleotide encoding saliva binding protein

| | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
| Image | | | | | | | | | |

KMC Draw Desc

28. Document ID: US 5798211 A

L2: Entry 28 of 47

File: USPT

Aug 25, 1998

US-PAT-NO: 5798211

DOCUMENT-IDENTIFIER: US 5798211 A

** See image for Certificate of Correction **

TITLE: Probe for diagnosing *Pseudomonas aeruginosa*

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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KMC Draw Desc

29. Document ID: US 5770375 A

L2: Entry 29 of 47

File: USPT

Jun 23, 1998

US-PAT-NO: 5770375

DOCUMENT-IDENTIFIER: US 5770375 A

** See image for Certificate of Correction **

TITLE: Probe for diagnosing *staphylococcus epidermidis*

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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KMC Draw Desc

30. Document ID: US 5763188 A

L2: Entry 30 of 47

File: USPT

Jun 9, 1998

US-PAT-NO: 5763188

DOCUMENT-IDENTIFIER: US 5763188 A

** See image for Certificate of Correction **

TITLE: Probe for diagnosing *Escherichia coli*, *Klebsiella pneumoniae* or *Enterobacter cloacae*

| | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments |
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| RRNA.USPT. | 2903 |
| RRNAS.USPT. | 307 |
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| (L1 AND (PROBE SAME RRNA)).USPT. | 47 |

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Search Results - Record(s) 31 through 40 of 47 returned.

31. Document ID: US 5723344 A

L2: Entry 31 of 47

File: USPT

Mar 3, 1998

US-PAT-NO: 5723344

DOCUMENT-IDENTIFIER: US 5723344 A

** See image for Certificate of Correction **

TITLE: Device for the capture of target molecules, and capturing process using the device

| | | | | | | | | | | | |
|--------------------------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
| <input type="button" value="Image"/> | | | | | | | | | | | |

32. Document ID: US 5721097 A

L2: Entry 32 of 47

File: USPT

Feb 24, 1998

US-PAT-NO: 5721097

DOCUMENT-IDENTIFIER: US 5721097 A

** See image for Certificate of Correction **

TITLE: Hybridization probes for the detection of branhamella catarrhalis strains

| | | | | | | | | | | | |
|--------------------------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
| <input type="button" value="Image"/> | | | | | | | | | | | |

33. Document ID: US 5703217 A

L2: Entry 33 of 47

File: USPT

Dec 30, 1997

US-PAT-NO: 5703217

DOCUMENT-IDENTIFIER: US 5703217 A

** See image for Certificate of Correction **

TITLE: Nucleotide fragment of the 23S ribosomal RNA of mycobacteria, derived probes and primers, reagent and detection method

| | | | | | | | | | | | |
|--------------------------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|------|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc |
| <input type="button" value="Image"/> | | | | | | | | | | | |

34. Document ID: US 5700928 A

L2: Entry 34 of 47

File: USPT

Dec 23, 1997

US-PAT-NO: 5700928
DOCUMENT-IDENTIFIER: US 5700928 A

TITLE: Polynucleotide encoding saliva binding protein

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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35. Document ID: US 5696251 A

L2: Entry 35 of 47

File: USPT

Dec 9, 1997

US-PAT-NO: 5696251
DOCUMENT-IDENTIFIER: US 5696251 A

TITLE: Non-nucleotide linking reagents for nucleotide probes

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

36. Document ID: US 5688646 A

L2: Entry 36 of 47

File: USPT

Nov 18, 1997

US-PAT-NO: 5688646
DOCUMENT-IDENTIFIER: US 5688646 A

TITLE: Mycoplasmas-agents for detecting and characterizing mycoplasmas in vitro

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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37. Document ID: US 5677123 A

L2: Entry 37 of 47

File: USPT

Oct 14, 1997

US-PAT-NO: 5677123
DOCUMENT-IDENTIFIER: US 5677123 A

TITLE: Mycoplasmas--agents for detecting and characterizing mycoplasmas in vitro

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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38. Document ID: US 5656744 A

L2: Entry 38 of 47

File: USPT

Aug 12, 1997

US-PAT-NO: 5656744
DOCUMENT-IDENTIFIER: US 5656744 A

TITLE: Methods for making nucleotide polymers using novel linking reagents

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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39. Document ID: US 5627030 A

L2: Entry 39 of 47

File: USPT

May 6, 1997

US-PAT-NO: 5627030

DOCUMENT-IDENTIFIER: US 5627030 A

TITLE: Method of amplification for increasing the sensitivity of detecting nucleic acid-probe target hybrids

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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40. Document ID: US 5585481 A

L2: Entry 40 of 47

File: USPT

Dec 17, 1996

US-PAT-NO: 5585481

DOCUMENT-IDENTIFIER: US 5585481 A

** See image for Certificate of Correction **

TITLE: Linking reagents for nucleotide probes

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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| PROBES.USPT. | 59010 |
| RRNA.USPT. | 2903 |
| RRNAS.USPT. | 307 |
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| (L1 AND (PROBE SAME RRNA)).USPT. | 47 |

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41. Document ID: US 5574145 A

L2: Entry 41 of 47

File: USPT

Nov 12, 1996

US-PAT-NO: 5574145

DOCUMENT-IDENTIFIER: US 5574145 A

TITLE: Isolated nucleic acid molecules targeted to the region intermediate to the 16S and 23S rRNA genes useful as probes for determining bacteria

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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42. Document ID: US 5571674 A

L2: Entry 42 of 47

File: USPT

Nov 5, 1996

US-PAT-NO: 5571674

DOCUMENT-IDENTIFIER: US 5571674 A

TITLE: DNA oligomers for use in detection of campylobacter pylori and methods of using such DNA oligomers

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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43. Document ID: US 5554517 A

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File: USPT

Sep 10, 1996

US-PAT-NO: 5554517

DOCUMENT-IDENTIFIER: US 5554517 A

TITLE: Nucleic acid amplification process

| | | | | | | | | | | | |
|-------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
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44. Document ID: US 5536638 A

L2: Entry 44 of 47

File: USPT

Jul 16, 1996

US-PAT-NO: 5536638

DOCUMENT-IDENTIFIER: US 5536638 A

** See image for Certificate of Correction **

TITLE: Hybridization probes derived from the spacer region between the 16S and 23S rRNA genes for the detection of *Neisseria gonorrhoeae*

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

45. Document ID: US 5409818 A

L2: Entry 45 of 47

File: USPT

Apr 25, 1995

US-PAT-NO: 5409818

DOCUMENT-IDENTIFIER: US 5409818 A

TITLE: Nucleic acid amplification process

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

46. Document ID: US 5378606 A

L2: Entry 46 of 47

File: USPT

Jan 3, 1995

US-PAT-NO: 5378606

DOCUMENT-IDENTIFIER: US 5378606 A

TITLE: Specific detection of *Neisseria gonorrhoeae*

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|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

47. Document ID: US 5173401 A

L2: Entry 47 of 47

File: USPT

Dec 22, 1992

US-PAT-NO: 5173401

DOCUMENT-IDENTIFIER: US 5173401 A

TITLE: Detection of *Neisseria gonorrhoeae*

| | | | | | | | | | | | |
|-----------------------|-------|----------|-------|--------|----------------|------|-----------|-----------|-------------|-----|-----------|
| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw Desc |
| Image | | | | | | | | | | | |

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| Term | Documents |
|----------------------------------|-----------|
| PROBE.USPT. | 111239 |
| PROBES.USPT. | 59010 |
| RRNA.USPT. | 2903 |
| RRNAS.USPT. | 307 |
| (1 AND (RRNA SAME PROBE)).USPT. | 47 |
| (L1 AND (PROBE SAME RRNA)).USPT. | 47 |

Display Format: TI Change Format

[Previous Page](#) [Next Page](#)

ds

| Set | Items | Description |
|---|-------|----------------------------|
| S1 | 0 | PY<1997 AND "2'-O-METHYL?" |
| S2 | 0 | "2'-O-METHYL" |
| S3 | 773 | PY<1997 AND ?METHYL? |
| S4 | 0 | S3 AND "2" (W) "O" |
| S5 | 4869 | PY<1997 AND HAIRPIN? |
| S6 | 172 | S5 AND METHYL? |
| S7 | 18 | S6 AND DETECT? |
| S8 | 14 | RD (unique items) |
| S9 | 162 | 2'-O-METHYL? |
| S10 | 156 | RD (unique items) |
| S11 | 0 | S5 AND S10 |
| S12 | 51 | S10 AND PY<1997 |
| S13 | 0 | S12 AND HAIRPIN? |
| S14 | 4 | S12 AND LOOP? |
| S15 | 6 | S12 NOT S14 AND STABL? |
| ? s s12 not s14 not s15 | | |
| | 51 | S12 |
| | 4 | S14 |
| | 6 | S15 |
| S16 | 41 | S12 NOT S14 NOT S15 |
| ? s s16 and oligonucleotide/ | | |
| >>>Possible typing error near end of line | | |
| ? s s16 and oligonucleotide? | | |
| | 41 | S16 |
| | 94019 | OLIGONUCLEOTIDE? |
| S17 | 5 | S16 AND OLIGONUCLEOTIDE? |
| ? t s17/3,ab/all | | |

17/3,AB/1 (Item 1 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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11168435 98044617 PMID: 9383369
Stabilizing effects of the RNA 2'-substituent: crystal structure of an oligodeoxynucleotide duplex containing 2'-O-methylated adenosines.
Lubini P; Zurcher W; Egli M
Organic Chemistry Laboratory, ETH Swiss Federal Institute of Technology, Zurich, Switzerland.
Chemistry & biology (ENGLAND) Sep 1994, 1 (1) p39-45, ISSN 1074-5521 Journal Code: 9500160
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
BACKGROUND: The stability of hybrids of 2'-O-methyl-ribonucleotides with complementary RNA is considerably higher than that of the corresponding DNA.RNA duplexes. The 2'-O-modified ribonucleotides are thus an attractive class of compounds for antisense applications. Understanding how these substituents stabilize the structure of the hybrid duplex may be important in the design of ribonucleotides with novel properties. RESULTS: The crystal structure of a dimer of the self-complementary DNA strand d(GCGT)O2'mer(A)d(TACGC), which has a 2'-O-methylated ribonucleotide incorporated at position 5, was determined at 2.1 Å resolution. This strand forms a duplex with an overall A-type conformation; the methyl groups of the two modified adenosines point into the relatively wide minor groove. Both 2'-methoxy groups are hydrogen-bonded to solvent molecules. These results allowed us to build a model of a fully 2'-O-methylated RNA double helix. CONCLUSIONS: Insertion of 2'-O-modified RNA residues into a stretch of DNA can nucleate a local A-type conformation, in part because modification with a bulky residue at this position stabilizes a C3'-endo type sugar pucker. The increased stability of fully 2'-O-methylated RNA may

several orders of magnitude lower than that of the tRNA-synthetase complex. Measurements of dissociation rate constants indicate that the stronger affinity of the 10-mer to tRNA-Cys is due to a significantly slower rate of dissociation (by a factor of 10-6) than that of the synthetase from the tRNA. Only a stoichiometric amount of the 10-mer is necessary to completely inhibit aminoacylation. Because tRNA aminoacylation is fundamental to cell growth, these results provide the rationale for the 10-mer and its derivatives as pharmaceutical agents that target specific cell growth.

1996

14/3,AB/2 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

10267039 BIOSIS NO.: 199698721957
NMR studies of a lead ribozyme and its non-cleavable analogue.
AUTHOR: Katahira Masato; Sugiyama Takashi; Kanagawa Mayumi; Kim Mi Hee; Uesugi Seichi(a); Kohno Toshiyuki
AUTHOR ADDRESS: (a)Dep. Bioengineering, Fac. Engineering, Yokohama National Univ., 156 Tokiwadai, Hodogaya-ku, Yokohama**Japan
JOURNAL: Nucleosides & Nucleotides 15 (1-3):p489-503 1996
ISSN: 0732-8311
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The structure of a lead ribozyme, which consists of two RNA strands, at neutral pH has been studied by NMR. Nearly all resonances of imino protons, base protons (H2, H5, H6 and H8) and sugar protons (H1' and H2') were assigned sequentially. Interesting structural features which deviate from the standard structure were found for the residues at an active site which consists of an internal loop. No indication of stable G:A base pairs was found in the loop. The effect of addition of Pb-2+ was studied by the use of a non-cleavable analogue in which the cytidine at a cleavage site is replaced by 2'-O-methylcytidine. It was suggested that Pb-2+ binds close to the cleavage-site and that the structural change induced by Pb-2+ is moderate and localized.

1996

14/3,AB/3 (Item 3 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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10143628 BIOSIS NO.: 199698598546
Selective binding of looped oligonucleotides to a single-stranded DNA and its influence on replication in vitro.
AUTHOR: Azhayeva Elena; Azhayev Alex(a); Guzaev Andrei; Lonnberg Harri
AUTHOR ADDRESS: (a)Dep. Chem., Univ. Turku, Turku, FIN-20500**Finland
JOURNAL: Nucleic Acids Research 23 (21):p4255-4261 1995
ISSN: 0305-1048
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Complexing of looped and circular oligonucleotides, composed of either 2'-deoxyribo- or 2'-O-methylribonucleoside units, with completely matching or partially mismatching complementary DNA sequences was studied. Melting experiments revealed considerable differences among the stabilities of these hybrid complexes. Maximum stability and

22/3,AB/17 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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10757930 BIOSIS NO.: 199799379075

Fluorescence resonance energy transfer analysis of the degradation of an oligonucleotide protected by a very **stable hairpin**.

AUTHOR: Refregiers Matthieu; Laigle Alain(a); Jolles Beatrice; Chinsky Laurent

AUTHOR ADDRESS: (a)Lab. Physicochimie Biomol. Cell., URA CNRS 2056,
Universite P. et M. Curie, Case 138, F-75252 Pa**France

JOURNAL: Journal of Biomolecular Structure & Dynamics 14 (3):p365-371

1996

ISSN: 0739-1102

RECORD TYPE: Abstract

LANGUAGE: English

X

ABSTRACT: In vitro degradation of **antisense** oligonucleotides protected or not on their 3' side against enzymatic attack by a naturally forming **hairpin** has been studied by fluorescence resonance energy transfer (FRET). The two oligonucleotides d(5'TTCTCGCGAAGC3') forming the **hairpin** and d(5'TTCTCCGGAAAGC3') as a control were labeled on their 5' side by tetramethylrhodamine and on their 3' side by fluorescein. Fluorescein has been shown not to hinder the **hairpin** formation and to give an additional protection against nucleases. The FRET technique proved adequate for an *in situ* study of these protected **antisense** oligonucleotides in living cells.

1996

22/3,AB/18 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

08269125 BIOSIS NO.: 000094050298

DIMERIZATION OF THE GENOMIC RNA OF HIV1 AND MoMuLV MECHANISM AND STRUCTURAL AND FUNCTIONAL IMPLICATIONS

AUTHOR: EHRESMANN B; MARQUET R; MOUGEL M; EHRESMANN C

AUTHOR ADDRESS: UPR CNRS 9002, STRUCTURE MACROMOLECULES BIOLOGIQUES

MECANISMES RECONNAISSANCE, INST. BIOLOGIE MOLECULAIRE CELLULAIRE, 67084 STRASBOURG CEDEX.

JOURNAL: BULL INST PASTEUR 90 (2). 1992. 109-124. 1992

FULL JOURNAL NAME: Bulletin de l'Institut Pasteur

CODEN: BIPAA

RECORD TYPE: Abstract

LANGUAGE: FRENCH

ABSTRACT: Dimerization is a key step in the retroviral cycle and is responsible for the high genetic recombination. It has also been proposed to positively regulate encapsidation and negatively, translation. In HIV1: (1) RNA fragments containing at least 100 nt down-stream from the 5' splice site can dimerize spontaneously; (2) dimerization is dependent on pH, temperature, RNA, mono- and multivalent cation concentrations, and the size of monovalent cations; (3) **antisense** RNA does not dimerize; (4) heterodimers can be formed between RNA from HIV1 and RNA from either MoMuLV or RSV. A consensus minimal polypurine RGGARA sequence has been identified in about 40 retroviruses and is present 4 times in HIV1 and twice in MoMuLV. The high thermal stability, cation size dependence and the low dissociation process led us to propose a mechanism involving purine quartets. In MoMuLV, region 215-312 is proposed to be required for dimerization. The rate of spontaneous dimerization depends on temperature and RNA and cation concentrations. Thermal denaturation

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07868656 93324340 PMID: 8392706

Self-stabilized antisense oligodeoxynucleotide phosphorothioates: properties and anti-HIV activity.

Tang J Y; Temsamani J; Agrawal S

Hybridon, Inc., Worcester, MA 01605.

Nucleic acids research (ENGLAND) Jun 11 1993, 21 (11) p2729-35

, ISSN 0305-1048 Journal Code: 0411011

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

A new class of oligodeoxyribonucleotides has been designed, referred to here as 'self-stabilized' oligonucleotides. These oligonucleotides have hairpin loop structures at their 3' ends, and show increased resistance to degradation by snake venom phosphodiesterase, DNA polymerase I and fetal bovine serum. The self-stabilized region of the oligonucleotide does not interfere in hybridization with complementary nucleic acids as shown by melting temperature, mobility-shift and RNase H cleavage studies. Various self-stabilized oligonucleotides containing increasingly stable hairpin loop regions were studied for their anti-HIV activity. Pharmacokinetic and stability studies in mice showed increased in vivo persistence of self-stabilized oligonucleotides with respect to their linear counterparts.

22/3,AB/14 (Item 14 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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07635541 93090736 PMID: 1280997

Implication of RNA structure on antisense oligonucleotide hybridization kinetics.

Lima W F; Monia B P; Ecker D J; Freier S M

Department of Molecular and Cellular Biology, Isis Pharmaceuticals, Carlsbad, California 92008.

Biochemistry (UNITED STATES) Dec 8 1992, 31 (48) p12055-61,

ISSN 0006-2960 Journal Code: 0370623

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

A 47-nucleotide transcript of the activated Ha-ras gene was prepared and determined, by enzymatic structure mapping, to form a stable hairpin structure. Six antisense decarboxylic nucleotides were designed, and association constants (K_a) for the hairpin - and length-matched complements were measured. Two of the antisense oligonucleotides targeted to the loop had nearly equal affinity for the transcript compared to the complement. The others, including one oligonucleotide complementary to the 3' side of the single-stranded loop, bound 10(5)-10(6)-fold less tightly to the transcript than to the short complement. We propose the difference in affinity is due to the target structure, both the secondary structure of the stem and the structure in the loop. Measurement of the bimolecular association rate constant, k_1 , and the dissociation rate constant, k_{-1} , for these oligonucleotides indicates the observed relationship between affinity and structure is primarily due to k_1 .

22/3,AB/15 (Item 15 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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*File 155: Medline has been reloaded and accession numbers have changed. Please see HELP NEWS 155.
File 5:Biosis Previews(R) 1969-2003/Mar W5
(c) 2003 BIOSIS
*File 5: Alert feature enhanced for multiple files, duplicates removal, customized scheduling. See HELP ALERT.

| Set | Items | Description |
|--|----------------------------|----------------------------|
| ? s | py<1997 and "2'-o-methyl?" | |
| Processing | | |
| 19971735 | PY<1997 | |
| 0 | 2'-O-METHYL? | |
| S1 | 0 | PY<1997 AND "2'-O-METHYL?" |
| ? s | "2'-o-methyl" | |
| S2 | 0 | "2'-O-METHYL" |
| ? s | py<1997 and ?methyl? | |
| >>>File 155 processing for ?METHYL? stopped at ALLMOGENS | | |
| >>>File 5 processing for ?METHYL? stopped at ACRT | | |
| 19971735 | PY<1997 | |
| 1206 | ?METHYL? | |
| S3 | 773 | PY<1997 AND ?METHYL? |
| ? s | s3 and "2" (w) "O" | |
| 773 | S3 | |
| 5399635 | 2 | |
| 401555 | O | |
| 18765 | 2 (W)O | |
| S4 | 0 | S3 AND "2" (W) "O" |
| ? s | py<1997 and hairpin? | |
| 19971735 | PY<1997 | |
| 9574 | HAIRPIN? | |
| S5 | 4869 | PY<1997 AND HAIRPIN? |
| ? s | s5 and methyl? | |
| >>>File 155 processing for METHYL? stopped at METHYLDINITROPHENOLS | | |
| >>>File 5 processing for METHYL? stopped at METHYLBUTOXYFURO | | |
| 4869 | S5 | |
| 467337 | METHYL? | |
| S6 | 172 | S5 AND METHYL? |
| ? s | s6 and detect? | |
| 172 | S6 | |
| 1615244 | DETECT? | |
| S7 | 18 | S6 AND DETECT? |
| ? rd | | |
| ...completed examining records | | |
| S8 | 14 | RD (unique items) |
| ? t | s8/3,ab/all | |

8/3,AB/1 (Item 1 from file: 155)
DIALOG(R)File 155: MEDLINE(R)
(c) format only 2003 The Dialog Corp. All rts. reserv.

10611669 96429277 PMID: 8832378
Homooligomeric dA.dU and dA.dT sequences in parallel and antiparallel strand orientation: consequence of the 5-methyl groups on stability, structure and interaction with the minor groove binding drug Hoechst 33258.
Germann M W; Kalisch B W; van de Sande J H
Jefferson Cancer Institute, Thomas Jefferson University, Philadelphia, PA
19107, USA. mwg@bern.jci.tju.edu
Journal of biomolecular structure & dynamics (UNITED STATES) Jun
1996, 13 (6) p953-62, ISSN 0739-1102 Journal Code: 8404176
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM

Record type: Completed

Oligodeoxyribonucleotides containing dA.dU base combinations were shown to form parallel stranded DNA. CD spectra and hyperchromicity profiles provide evidence that the structure is very similar to that of a related parallel stranded dA.dT oligomer. Thermal denaturation studies show that these parallel dA.dU sequences are significantly less stable than their dA.dT analogues in either antiparallel or parallel stranded orientations. The stabilizing effect of the 5-methyl group is similar for parallel and antiparallel sequences. The minor groove binding drug Hoechst 33258 binds with similar affinity to APS dA.dT and APS dA.dU sequences. However, binding to the PS dA.dT hairpin is significantly impaired as a consequence of the different groove dimensions and the presence of thymine methyl groups at the binding site. This results in an 8.6 kJmol-1 reduced free energy of binding for the PS dA.dT sequence. Replacement of the bulky methyl group with a hydrogen (ie. T-->U) results in significantly stronger Hoechst 33258 binding to the parallel dA.dU sequences with a penalty of only 4.1 kJmol-1. Our data demonstrate that although Hoechst 33258 detects the altered groove, it is still able to bind a PS duplex containing dA.dU base pairs with high affinity, despite the large structural differences from its regular binding site in APS DNA.

8/3,AB/2 (Item 2 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08707256 95395861 PMID: 7666435

DNA palindromes adopt a methylation-resistant conformation that is consistent with DNA cruciform or hairpin formation in vivo.

Allers T; Leach D R

Institute of Cell and Molecular Biology, University of Edinburgh, UK.

Journal of molecular biology (ENGLAND) Sep 8 1995, 252 (1)

p70-85, ISSN 0022-2836 Journal Code: 2985088R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Long DNA palindromes present a threat to genomic stability and are not tolerated in *Escherichia coli*. It has been suggested that this is a consequence of cruciform or hairpin formation by palindromic sequences. This work describes a methylation inhibition assay for unusual DNA secondary structure in vivo that is both internally controlled and non-invasive. If a palindrome with a central GATC target site for Dam methylase assumes a cruciform or hairpin conformation in vivo, then the GATC sequence will be located in a single-stranded loop and will consequently not be modified. The centre of a long perfect palindrome located in bacteriophage lambda is shown to be methylation-resistant in vivo. Changes to the central sequence and insertions of 10 base-pairs of asymmetric sequence do not alter the degree of under-methylation, but insertions of 20 base-pairs or more of asymmetric sequence reduce the under-methylation of the palindrome centre. We also show that the centres of long palindromes are more under-methylated than equivalent sequences in a non-palindromic context. These results are consistent with an unusual secondary structure, such as DNA cruciform or hairpin, and indicate that the formation pathway of the structure detected is independent of the composition and symmetry of the central 10 base-pairs of the palindrome.

8/3,AB/3 (Item 3 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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07797410 93252944 PMID: 8486707

EBP-80, a transcription factor closely resembling the human autoantigen Ku, recognizes single- to double-strand transitions in DNA.

Falzon M; Fewell J W; Kuff E L

Laboratory of Biochemistry, National Cancer Institute, Bethesda, Maryland 20892.

Journal of biological chemistry (UNITED STATES) May 15 1993, 268 (14) p10546-52, ISSN 0021-9258 Journal Code: 2985121R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

We have previously reported the purification and characterization of the transcription factor EBP-80 (Falzon, M., and Kuff, E. L. (1989) J. Biol. Chem. 264, 21915-21922). EBP-80 mediates the DNA methylation effect on transcription from an endogenous proviral long terminal repeat. Here we show that EBP-80 is very similar if not identical to the Ku autoantigen, a heterodimeric nuclear protein first detected by antibodies from autoimmune patients (Mimori, T., Akizuki, M., Yamagata, H., Inada, S., Yoshida, S., and Homma, M. (1981) J. Clin. Invest. 68, 611-620). A number of laboratories have shown that the Ku protein complex binds to free double-stranded DNA ends. In this study, we have examined the binding properties of EBP-80. EBP-80 binds single-stranded DNA with low affinity. Binding to random sequence double-stranded DNA depends on the length of the duplex and is optimal with oligomers of 30 and 32 base pairs; the protein complexes formed with these oligomers have Kd values of 15-20 pM. It binds with comparable high affinities to blunt-ended duplex DNA, to duplex DNA ending in hairpin loops, and to constructs in which an internal segment of duplex DNA is flanked by single-strand extensions. EBP-80 also interacts effectively with circular duplex molecules containing a 30-nucleotide single-stranded region (gap) or a double-stranded segment of nonhomology (bubble), but only weakly with the corresponding closed circular construct made up entirely of duplex DNA. EBP-80 prefers A/T to G/C ends. The binding properties of EBP-80 are consistent with the hypothesis that it recognizes single- to double-strand transitions in DNA. A model is presented for the interaction of EBP-80 with its target sequence in the proviral long terminal repeat.

8/3,AB/4 (Item 4 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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07653246 93108447 PMID: 1469716

Structure analysis of the 5' external transcribed spacer of the precursor ribosomal RNA from *Saccharomyces cerevisiae*.

Yeh L C; Lee J C

Department of Biochemistry, University of Texas Health Science Center, San Antonio 78284-7760.

Journal of molecular biology (ENGLAND) Dec 5 1992, 228 (3) p827-39, ISSN 0022-2836 Journal Code: 2985088R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Full-length precursor ribosomal RNA molecules were produced in vitro using as a template, a plasmid containing the yeast 35 S pre-rRNA gene under the control of the phage T3 promoter. The higher-order structure of the 5'-external transcribed spacer (5' ETS) sequence in the 35S pre-rRNA molecule was studied using dimethylsulfate, 1-cyclohexyl-3-(2-morpholinoethyl)-carbodiimide metho-p-toluenesulfonate, RNase T1 and RNase V1 as structure-sensitive probes. Modified residues were detected by primer extension. Data produced were used to evaluate several theoretical structure models predicted by minimum free-energy calculations. A model for the entire 5'ETS region is proposed that accommodates 82% of the residues

experimentally shown to be in either base-paired or single-stranded structure in the correct configuration. The model contains a high degree of secondary structure with ten stable hairpins of varying lengths and stabilities. The hairpins are composed of the Watson-Crick A.T and G.C pairs plus the non-canonical G.U pairs. Based on a comparative analysis of the 5' ETS sequence from *Saccharomyces cerevisiae* and *Schizosaccharomyces pombe*, most of the base-paired regions in the proposed model appear to be phylogenetically supported. The two sites previously shown to be crosslinked to U3 snRNA as well as the previously proposed recognition site for processing and one of the early processing site (based on sequence homology to the vertebrate ETS cleavage site) are located in single-stranded regions in the model. The present folding model for the 5' ETS in the 35 S pre-rRNA molecule should be useful in the investigations of the structure, function and processing of pre-rRNA.

8/3,AB/5 (Item 5 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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07620507 93075690 PMID: 1445839

The role of 5-methylcytidine in the anticodon arm of yeast tRNA(Phe): site-specific Mg²⁺ binding and coupled conformational transition in DNA analogs.

Dao V; Guenther R H; Agris P F
Department of Biochemistry, North Carolina State University, Raleigh 27695.

Biochemistry (UNITED STATES) Nov 17 1992, 31 (45) p11012-9,
ISSN 0006-2960 Journal Code: 0370623

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

The tDNA(Phe)AC, d(CCAGACTGAAGAU13m5C14U15GG), with a DNA sequence similar to that of the anticodon stem and loop of yeast tRNA(Phe), forms a stem and loop structure and has an Mg(2+)-induced structural transition that was not exhibited by an unmodified tDNA(Phe)AC d(T13C14T15) [Guenther, R. H., Hardin, C. C., Sierzputowska-Gracz, H., Dao, V., & Agris, P. F. (1992) Biochemistry (preceding paper in this issue)]. Three tDNA(Phe)AC molecules having m5C14, tDNA(Phe)AC d(U13m5C14U15), d(U13m5C14T15), and d(T13,5C14U15), also exhibited Mg(2+)-induced structural transitions and biphasic thermal transitions (T_m approximately 23.5 and 52 degrees C), as monitored by CD and UV spectroscopy. Three other tDNA(Phe)AC, d(T13C14T15), d(U13C14U15), and d(A7;U13m5C14U15) in which T7 was replaced with an A, thereby negating the T7.A10 base pair across the anticodon loop, had no Mg(2+)-induced structural transitions and only monophasic thermal transitions (T_m of approximately 52 degrees C). The tDNA(Phe)AC d(U13m5C14U15) had a single, strong Mg²⁺ binding site with a K_d of 1.09 x 10(-6) M and a delta G of -7.75 kcal/mol associated with the Mg(2+)-induced structural transition. In thermal denaturation of tDNA(Phe)AC d(U13m5C14U15), the 1H NMR signal assigned to the imino proton of the A5.dU13 base pair at the bottom of the anticodon stem could no longer be detected at a temperature corresponding to that of the loss of the Mg(2+)-induced conformation from the CD spectrum. Therefore, we place the magnesium in the upper part of the tDNA hairpin loop near the A5.dU13 base pair, a location similar to that in the X-ray crystal structure of native, yeast tRNA(Phe). (ABSTRACT TRUNCATED AT 250 WORDS)

8/3,AB/6 (Item 6 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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06678224 90304111 PMID: 1694683

On the chemical nature of DNA and RNA modification by a hemin model system.

Van Atta R B; Bernadou J; Meunier B; Hecht S M

Department of Chemistry, University of Virginia, Charlottesville 22901.

Biochemistry (UNITED STATES) May 22 1990, 29 (20) p4783-9,

ISSN 0006-2960 Journal Code: 0370623

Contract/Grant No.: AI27185; AI; NIAID

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

In order to model the interaction of hemin with DNA and other polynucleotides, we have studied the degradation of DNA, RNA, and polynucleotides of defined structure by [meso-tetrakis(N-methyl-4-pyridyl)porphinato]manganese(III) (MnTMPP) + KHSO5. The activated porphyrin was shown to release adenine, thymine, and cytosine from DNA; RNA degradation afforded adenine, uracil, and cytosine. The same products were obtained from single- and double-stranded DNA oligonucleotides of defined sequence, and also from single-stranded DNA and RNA homopolymers. The overall yield of bases from the dode-canucleotide d(CGCT3A3GCG) was equal to 14% of the nucleotides present initially, indicating that each porphyrin catalyzed the release of approximately 4 bases. Although no guanine was detected as a product from any of the substrates studied, the ability of MnTMPP + KHSO5 to degrade guanine nucleotides was verified by the destruction of pGp, and by the appearance of bands corresponding to guanosine cleavage following treatment of 32P end labeled DNA restriction fragments with activated MnTMPP. Inspection of a number of sites of MnTMPP-promoted cleavage indicated that the process was sequence-selective, occurring primarily at G residues that were part of 5'-TG-3' or 5'-AG-3' sequences, or at T residues. Also formed in much greater abundance were alkali-labile lesions; these were formed largely at guanosine residues. Also studied was the degradation of a 47-nucleotide RNA molecule containing two hairpins. Degradation of the 5'-32P end labeled RNA substrate afforded no distinct, individual bands, suggesting that multiple modes of degradation may be operative. (ABSTRACT TRUNCATED AT 250 WORDS)

8/3,AB/7 (Item 7 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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06587341 90212664 PMID: 2322582

Comparative analysis of 5.8 S rRNA from Ephedra kakanica Regel. (Gymnospermae) and other plant species.

Melekhovets YuF; Troitsky A V

A.N. Belozersky Laboratory of Molecular Biology and Bioorganic Chemistry, Moscow State University, U.S.S.R.

Biochimica et biophysica acta (NETHERLANDS) Apr 6 1990, 1048 (2-3) p294-6, ISSN 0006-3002 Journal Code: 0217513

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

5.8 S rRNA from the gymnosperm Ephedra kakanica Regel. (EMBL Data Library accession No. X15676) has been sequenced. It is 161 nucleotides long and contains three 2'-O-methylated residues--two adenosines and one guanosine. No pseudouridine have been detected. E. kakanica 5.8 S rRNA, as those from other plant species, can form a secondary structure with paired 5'- and 3'-terminal regions. 5.8 S rRNAs of seed plants differ from the moss Mnium reguicum 5.8 S rRNA in that they have longer variable 'GC-rich' hairpins with insertions in the loop region. 5.8 S rRNA of E. kakanica reveals 69 and 82% of homology with that of moss and five angiosperm species, respectively. The posttranscriptional modification pattern of plant 5.8 S rRNAs is not strictly conservative.

8/3,AB/8 (Item 8 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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06243265 89259096 PMID: 2542611
Identification of nuclear proteins that specifically interact with adeno-associated virus type 2 inverted terminal repeat hairpin DNA.
Ashktorab H; Srivastava A
Department of Medicine, Indiana University School of Medicine, Indianapolis 46202.
Journal of virology (UNITED STATES) Jul 1989, 63 (7) p3034-9,
ISSN 0022-538X Journal Code: 0113724
Contract/Grant No.: AI-26323; AI; NIAID
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
A palindromic hairpin duplex containing the inverted terminal repeat sequence of adeno-associated virus type 2 (AAV) DNA was used as a substrate in gel retardation assays to detect putative proteins that specifically interact with the AAV hairpin DNA structures. Nuclear proteins were detected in extracts prepared from human KB cells coinfecte with AAV and adenovirus type 2 that interacted with the hairpin duplex but not in nuclear extracts prepared from uninfected, AAV-infected, or adenovirus type 2-infected KB cells. The binding was specific for the hairpin duplex, since no binding occurred with a double-stranded DNA duplex with the identical nucleotide sequence. Furthermore, in competition experiments, the binding could be reduced with increasing concentrations of the hairpin duplex but not with the double-stranded duplex DNA with the identical nucleotide sequence. S1 nuclease assays revealed that the binding was sensitive to digestion with the enzyme, whereas the protein-bound hairpin duplex was resistant to digestion with S1 nuclease. The nucleotide sequence involved in the protein binding was localized within the inverted terminal repeat of the AAV genome by methylation interference assays. These nuclear proteins may be likely candidates for the pivotal enzyme nuclease required for replication or resolution (or both) of single-stranded palindromic hairpin termini of the AAV genome.

8/3,AB/9 (Item 9 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
(c) format only 2003 The Dialog Corp. All rts. reserv.

03604334 82014865 PMID: 6169000
Primary and secondary structures of chicken, rat and man nuclear U4 RNAs.
Homologies with U1 and U5 RNAs.
Krol A; Brannan C; Lazar E; Gallinaro H; Jacob M
Nucleic acids research (ENGLAND) Jun 25 1981, 9 (12) p2699-716
ISSN 0305-1048 Journal Code: 0411011
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
U4 RNA from chicken, rat and man was examined for nucleotide sequence and secondary structure. Three molecular species, U4A, U4B and U4C were detected in the three animal species. U4A is 146 nucleotide long and U4B RNA only lacks the 3' terminal G. four nucleotides are missing at the 3'-end of U4C RNA which, in addition, differs from U4A and U4B RNAs at two internal positions. Thus, U4C RNA is encoded by another gene as U4A and U4B RNAs. Only one nucleotide substitution occurred between chicken and man showing that U4A, U4B and U4C RNAs have been extremely conserved throughout

evolution. The three molecular species are capped, they contain three psi, a 2'-P methyl A and a m6A. An additional post-transcriptional modification close to the cap structure is observed in man. On the basis on an experimental study, two models of secondary structure may be proposed for U4 RNA. The 3' domain is the same in both models and is homologous to that of U1 and U5 RNAs. It consists of a single-stranded region, containing the sequence Py-(A)2-(U)n-Gp flanked by two stable hairpins probably involved in tertiary interactions. The 5' domain is less stable than the 3' domain and its structure is different in the two models. However, a long single-stranded pyrimidine region containing modified nucleotides is found in both models as in U1 and U5 RNAs. Several other nucleotide sequence homologies related to specific features of secondary structure suggest that U1, U4 and U5 RNAs derive from a common ancestor and may have common function.

8/3,AB/10 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

10508103 BIOSIS NO.: 199699129248

Homooligomeric dA cndot dU and dA cndot dT sequences in parallel and antiparallel strand orientation: Consequence of the 5-methyl groups on stability, structure and interaction with the minor groove binding drug HOECHST 33258.

AUTHOR: Germann Markus W(a); Kalisch Bernd W; Van De Sande Johan H

AUTHOR ADDRESS: (a)Jefferson Cancer Inst., Dep. Pharmacology, Thomas Jefferson Univ., 233 South 10 Street, Philadelphia USA

JOURNAL: Journal of Biomolecular Structure & Dynamics 13 (6):p953-962

1996

ISSN: 0739-1102

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: Oligodeoxyribonucleotides containing dA cndot dU base combinations were shown to form parallel stranded DNA. CD spectra and hyperchromicity profiles provide evidence that the structure is very similar to that of a related parallel stranded dA cndot dT oligomer. Thermal denaturation studies show that these parallel dA cndot dU sequences are significantly less stable than their dA cndot dT analogues in either antiparallel or parallel stranded orientations. The stabilizing effect of the 5-methyl group is similar for parallel and antiparallel sequences. The minor groove binding drug Hoechst 33258 binds with similar affinity to APS dA cndot dT and APS dA cndot dU sequences. However, binding to the PS dA cndot dT hairpin is significantly impaired as a consequence of the different groove dimensions and the presence of thymine methyl groups at the binding site. This results in an 8.6 kJ mol-1 reduced free energy of binding for the PS dA cndot dT sequence. Replacement of the bulky methyl group with a hydrogen (ie. T fwdarw U) results in significantly stronger Hoechst 33258 binding to the parallel dA cndot dU sequences with a penalty of only 4.1 kJ mol-1. Our data demonstrate that although Hoechst 33258 detects the altered groove, it is still able to bind a PS duplex containing dA cndot dU base pairs with high affinity, despite the large structural differences from its regular binding site in APS DNA.

1996

8/3,AB/11 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

08887971 BIOSIS NO.: 199396039472

Excision of 3-methylguanine from alkylated DNA by 3-methyladenine DNA glycosylase I of Escherichia coli.

AUTHOR: Bjelland Svein(a); Bjoras Magnar; Seeberg Erling

AUTHOR ADDRESS: (a)Norwegian Defence Res. Establishment, Div. Environ. Toxicol., P.O. Box 25, N-2007 Kjeller**Norway

JOURNAL: Nucleic Acids Research 21 (9):p2045-2049 1993

ISSN: 0305-1048

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: Escherichia coli has two DNA glycosylases for repair of DNA damage caused by simple alkylating agents. The inducible AlkA DNA glycosylase (3-methyladenine (m-3A) DNA glycosylase II) removes several different alkylated bases including m-3A and 3-methylguanine (m-3G) from DNA, whereas the constitutively expressed Tag enzyme (m-3A DNA glycosylase I) has appeared to be specific for excision of M-3A. In this communication we have reexamined the substrate specificity of Tag by using synthetic DNA rich in GC base pairs to facilitate **detection** of any possible methyl-G removal. In such DNA alkylated with (3H)dimethyl sulphate, we found that m-3G was excised from double-stranded DNA by both glycosylases, although more efficiently by AlkA than by Tag. This was further confirmed using both N-(3H) methyl-N-nitrosourea- and (3H)dimethyl sulphate-treated native DNA, from which Tag excised m-3G with an efficiency that was about 70 times lower than for AlkA. These results can explain the previous observation that high levels of Tag expression will suppress the alkylation sensitivity of alkA mutant cells, further implying that m-3G is formed in quantity sufficient to represent an important cytotoxic lesion if left unrepaired in cells exposed to alkylating agents.

1993

8/3,AB/12 (Item 3 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

(c) 2003 BIOSIS: All rts. reserv.

07255625 BIOSIS NO.: 000090035501

THE REFINED 2.4 A X-RAY CRYSTAL STRUCTURE OF RECOMBINANT HUMAN STEFIN B IN COMPLEX WITH THE CYSTEINE PROTEINASE PAPAIN A NOVEL TYPE OF PROTEINASE INHIBITOR INTERACTION

AUTHOR: STUBBS M T; LABER B; BODE W; HUBER R; JERALA R; LENARCIC B; TURK V

AUTHOR ADDRESS: MAX-PLANCK-INST. BIOCHEMIE, D-8033 MARTINSRIED BEI MUENCHEN, WEST GERMANY.

JOURNAL: EMBO (EUR MOL BIOL ORGAN) J 9 (6). 1990. 1939-1948. 1990

FULL JOURNAL NAME: EMBO (European Molecular Biology Organization) Journal

CODEN: EMJOD

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

ABSTRACT: A stoichiometric complex of human stefin B and carboxy methylated papain has been crystallized in a trigonal crystal form. Data to 2.37 .ANG. resolution were collected using the area detector diffractometer FAST. The crystal structure of the complex has been solved by Patterson search techniques using papain as search model. Starting from the structure of chicken cystatin, the stefin structure was elucidated through cycles of model building and crystallographic refinement. The current crystallographic R factor is 0.19. Like cystatin, the stefin molecule consists of a five stranded .beta.-sheet wrapped around a five turn .alpha.-helix, but with an additional carboxy terminal strand running along the convex side of the sheet. Topological equivalence of stefin and cystatin reveal the previous

sequence alignment to be incorrect in part, through deletion of the intermediate helix. The conserved residues form a tripartite wedge, which slots into the papain active site as proposed through consideration of the tertiary structures of the individual components (Bode et al., 1988). The main interactions are provided by the amino terminal 'trunk' (occupying the 'unprimed' subsites of the enzyme), and by the first hairpin loop, containing the highly conserved QVVAG sequence, with minor contributions from the second hairpin loop. The carboxyl terminus of stefin provides an additional interaction region with respect to cystatin. The interaction is dominated by hydrophobic contacts. Inhibition by the cysteine proteinase inhibitors is fundamentally different to that observed for the serine proteinase inhibitors.

1990

8/3,AB/13 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

07235629 BIOSIS NO.: 000090015503
COMPARATIVE ANALYSIS OF 5.8S RIBOSOMAL RNA FROM EPHEDRA-KOKANICA REGL.

GYMNOSPERMAE AND OTHER PLANT SPECIES

AUTHOR: MELEKHOVETS Y F; TROITSKY A V

AUTHOR ADDRESS: A.N. BELOZERSKY LABORATORY MOLECULAR BIOL. BIOORGANIC CHEMISTRY, MOSCOW STATE UNIV., MOSCOW 119899, U.S.S.R.

JOURNAL: BIOCHIM BIOPHYS ACTA 1048 (2-3). 1990. 294-296. 1990

FULL JOURNAL NAME: Biochimica et Biophysica Acta

CODEN: BBACA

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

ABSTRACT: 5.8 S rRNA from the gymnosperm Ephedra kakanica Regl. (EMBL Data Library accession No. X15676) has been sequenced. It is 161 nucleotides long and contains three 2'-O-methylated residues.sbd.two adenosines and one guanosine. No pseudouridine have been detected. E. kakanica 5.8 S rRNA, as those from other plant species, can form a secondary structure with paired 5'- and 3'-terminal regions. 5.8 S rRNAs of seed plants differ from the moss Mnium rugicum 5.8 S rRNA in that they have longer variable 'GC-rich' hairpins with insertions in the loop region. 5.8 S rRNA of E. kakanica reveals 69 and 82% of homology with that of moss and five angiosperm species, respectively. The posttranscriptional modification pattern of plant 5.8 S rRNAs is not strictly conservative.

1990

8/3,AB/14 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

06647038 BIOSIS NO.: 000087089215

7 DEAZA-2'-DEOXY-O-6-METHYLGUANOSINE SELECTIVE N-2 FORMYLATION VIA A FORMAMIDINE PHOSPHORAMIDITE SYNTHESIS AND PROPERTIES OF OLIGONUCLEOTIDES

AUTHOR: SEELA F; DRILLER H

AUTHOR ADDRESS: LAB. ORG. BIOORG. CHEM., FACHBEREICH BIOL./CHEM., UNIV. OSNABRUECK, D-4500 OSNABRUECK, WEST-GERMANY.

JOURNAL: NUCLEOSIDES NUCLEOTIDES 8 (1). 1989. 1-22. 1989

FULL JOURNAL NAME: Nucleosides & Nucleotides

CODEN: NUNUD

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

ABSTRACT: Hexa- and dodecanucleotides containing 7-deaza-2'-deoxy-06-methylguanosine (m6c7Gd, 2) have been prepared by solid-phase synthesis employing methyl- as well as cyanoethyl phosphoramidites of 2. As the N2-isobuturyl group of protected 2 was difficult to remove after oligonucleotide synthesis the more labile formyl group was introduced. Regioselective N2-formylation was carried out without sugar protection by in-situ hydrolysis of a formamidine intermediate. replacement of dG within the oligomer d(CGCGAATTCGCG) next to dA increased hairpin formation, whereas duplexes were formed if one of the outer dG residues was replaced by m6c7Gd. Tertiary structure changes of the oligomers were detected by comparing the melting curves at 260 and 280 nm or duplex specific cleavage by the endodeoxyribonuclease Eco RI.

1989

```

s 2'-O-methyl?
>>>Warning: unmatched quote found
      S9      162  2'-O-METHYL?
? rd
...examined 50 records (50)
...examined 50 records (100)
...examined 50 records (150)
...completed examining records
      S10      156  RD (unique items)
? ds

Set      Items      Description
S1      0      PY<1997 AND "2'-O-METHYL?"
S2      0      "2'-O-METHYL"
S3      773     PY<1997 AND ?METHYL?
S4      0      S3 AND "2" (W) "O"
S5      4869    PY<1997 AND HAIRPIN?
S6      172      S5 AND METHYL?
S7      18       S6 AND DETECT?
S8      14       RD (unique items)
S9      162     2'-O-METHYL?
S10     156     RD (unique items)
? s s5 and s10
      4869  S5
      156   S10
      S11     0  S5 AND S10
? s s10 and py<1997
      156  S10
      19971735 PY<1997
      S12     51  S10 AND PY<1997
? s s12 and hairpin?
      51   S12
      9574    HAIRPIN?
      S13     0  S12 AND HAIRPIN?
? s s12 and loop?
      51   S12
      116568  LOOP?
      S14     4  S12 AND LOOP?
? t s14/3,ab/all

```

14/3,AB/1 (Item 1 from file: 5)
 DIALOG(R) File 5:Biosis Previews(R)
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10713662 BIOSIS NO.: 199799334807
 Inhibition of tRNA aminoacylation by 2'-O-methyl oligonucleotides.
 AUTHOR: Hou Ya-Ming(a); Gamper Howard B
 AUTHOR ADDRESS: (a)Dep. Biochem. Mol. Pharmacol., Thomas Jefferson Univ.,
 233 S. 10th St., Philadelphia, PA 19107**USA
 JOURNAL: Biochemistry 35 (48):p15340-15348 1996
 ISSN: 0006-2960
 RECORD TYPE: Abstract
 LANGUAGE: English

ABSTRACT: A 2'-O-methyl oligonucleotide complementary to 18 nucleotides in the dihydrouridine stem-loop of *Escherichia coli* tRNA-Cys, has been shown to stably bind to the tRNA. The binding inhibits aminoacylation of the tRNA by cysteine tRNA synthetase. The same oligonucleotide sequence but with the DNA deoxy backbone does not bind to the tRNA. This provides the basis for the design and test of a series of 2'-O-methyl oligonucleotides for their ability to bind to *E. coli* tRNA-Cys and inhibit aminoacylation. We show here that different regions of the tRNA have different sensitivities to oligonucleotides. A 10-mer that targets G15 forms a stable complex with the tRNA. The K-d of the complex is

selectivity was displayed by oligomers 2 and 5. It was concluded that a linear stretch, attached to 1'-O- of 3'-deoxypsicothymidine unit (Z) increases the selectivity of hybridization and stability of the complex as a whole. This allows one to aim the target DNA very precisely at its polyadenine part as well as at adjacent sequence simultaneously. Experiments on termination of primer extension catalysed by different DNA-polymerases-Sequenase, Klenow fragment and Tth-have demonstrated that **looped** oligomer 5, composed of 2'-O-methylribonucleosides appears to be a highly selective and potent inhibitor of replication in vitro. Features of **looped** oligonucleotides, composed of 2'-O-methylribonucleosides seem to be useful for design of highly specific antigenic oligonucleotides.

1995

14/3,AB/4 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
(c) 2003 BIOSIS. All rts. reserv.

09649270 BIOSIS NO.: 199598104188
Modified nucleosides and codon recognition.
BOOK TITLE: tRNA: Structure, biosynthesis, and function
AUTHOR: Yokoyama Shigeyuki (a); Nishimura Susumu
BOOK AUTHOR/EDITOR: Soell D; RajBhandary U L: Eds
AUTHOR ADDRESS: (a)Dep. Biophysics Biochem., Sch. Sci., Univ. Tokyo, 7-3-1
Hongo, Bunkyo-ku, Tokyo 113**Japan
p207-223 1995
BOOK PUBLISHER: American Society for Microbiology (ASM), Books Division,
1325 Massachusetts Ave. NW, Washington, DC 20005-4171,
USA
ISBN: 1-55581-073-X
DOCUMENT TYPE: Book
RECORD TYPE: Citation
LANGUAGE: English
1995
?

ds

| Set | Items | Description |
|----------------------------|--------|----------------------------|
| S1 | 0 | PY<1997 AND "2'-O-METHYL?" |
| S2 | 0 | "2'-O-METHYL" |
| S3 | 773 | PY<1997 AND ?METHYL? |
| S4 | 0 | S3 AND "2" (W) "O" |
| S5 | 4869 | PY<1997 AND HAIRPIN? |
| S6 | 172 | S5 AND METHYL? |
| S7 | 18 | S6 AND DETECT? |
| S8 | 14 | RD (unique items) |
| S9 | 162 | 2'-O-METHYL? |
| S10 | 156 | RD (unique items) |
| S11 | 0 | S5 AND S10 |
| S12 | 51 | S10 AND PY<1997 |
| S13 | 0 | S12 AND HAIRPIN? |
| S14 | 4 | S12 AND LOOP? |
| ? s s12 not s14 and stabl? | | |
| | 51 | S12 |
| | 4 | S14 |
| | 353726 | STABL? |
| S15 | 6 | S12 NOT S14 AND STABL? |
| ? t s15/3,ab/all | | |

15/3,AB/1 (Item 1 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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08373440 95061413 PMID: 7526343
Interactions of oligonucleotide analogs containing methylphosphonate internucleotide linkages and 2'-O-methylribonucleosides.
Kean J M; Cushman C D; Kang H; Leonard T E; Miller P S
Department of Biochemistry, School of Hygiene and Public Health, Johns Hopkins University, Baltimore, MD 21205.
Nucleic acids research (ENGLAND) Oct 25 1994, 22 (21)
p4497-503, ISSN 0305-1048 Journal Code: 0411011
Contract/Grant No.: CA42762; CA; NCI
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The interactions of oligonucleotide analogs, 12-mers, which contain deoxyribo- or 2'-O-methylribose sugars and methylphosphonate internucleotide linkages with complementary 12-mer DNA and RNA targets and the effect of chirality of the methylphosphonate linkage on oligomer-target interactions was studied. Oligomers containing a single Rp or Sp methylphosphonate linkage (type 1) or oligomers containing a single phosphodiester linkage at the 5'-end followed by 10 contiguous methylphosphonate linkages of random chirality (type 2) were prepared. The deoxyribo- and 2'-O-methylribo- type 1 12-mers formed **stable** duplexes with both the RNA and DNA as determined by UV melting experiments. The melting temperatures, Tms, of the 2'-O-methylribo-12-mer/RNA duplexes (49-53 degrees C) were higher than those of the deoxyribo-12mer/RNA duplexes (31-36 degrees C). The Tms of the duplexes formed by the Rp isomers of these oligomers were approximately 3-5 degrees C higher than those formed by the corresponding Sp isomers. The deoxyribo type 2 12-mer formed a **stable** duplex, Tm 34 degrees C, with the DNA target and a much less **stable** duplex with the RNA target, Tm < 5 degrees C. In contrast, the 2'-O-methylribo type 2 12-mer formed a **stable** duplex with the RNA target, Tm 20 degrees C, and a duplex of lower stability with the DNA target, Tm < 5 degrees C. These results show that the previously observed greater stability of oligo-2'-O-methylribonucleotide/RNA duplexes versus oligodeoxyribonucleotide/RNA duplexes extends to oligomers containing methylphosphonate linkages and that the configuration of the

methylphosphonate linkage strongly influences the stability of the duplexes.

15/3,AB/2 (Item 2 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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07475954 92339503 PMID: 1633847
Oligo(2'-O-methyl)ribonucleotides. Effective probes for duplex DNA.
Shimizu M; Konishi A; Shimada Y; Inoue H; Ohtsuka E
Faculty of Pharmaceutical Sciences, Hokkaido University, Sapporo, Japan.
FEBS letters (NETHERLANDS) May 11 1992, 302 (2) p155-8, ISSN
0014-5793 Journal Code: 0155157
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
To find novel probes for duplex DNA, we prepared four types of triplexes containing a homopurine-homopyrimidine 15-mer duplex DNA, and examined their thermal stabilities (T_m values). The single strand used for triplex formation were a DNA 15-mer having a defined C-T mixed sequence, and its sugar-modified analogs, namely 2'-fluoro DNA, RNA, and 2'-O-methyl RNA. The 2'-O-methyl RNA and the RNA-containing triplexes were similar in their enhanced stabilities at pH 6.1 and, amongst the four triplexes, the 2'-O-methyl was the most **stable** at pH 5.0. Furthermore, an experiment using a 34-mer duplex DNA suggested that the 2'-O-methyl RNA-triplex was destabilized, mostly as a result of the incorporation of a mismatched triplet, as compared to the DNA triplex counterpart. Thus, 2'-O-methyl RNA can serve as an effective probe for duplex DNA.

15/3,AB/3 (Item 1 from file: 5)
DIALOG(R) File 5:Biosis Previews(R)
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10481575 BIOSIS NO.: 199699102720
Intron-encoded, antisense small nucleolar RNAs: The characterization of nine novel species points to their direct role as guides for the 2'-O-ribose methylation of rRNAs.
AUTHOR: Nicoloso Monique; Qu Liang-Hu; Michot Bernard; Bachellerie Jean-Pierre(a)
AUTHOR ADDRESS: (a)Lab. Biol. Mol. Eucaryote C.N.R.S., Univ. Paul-Sabatier,
118 route Narbonne, 31062 Toulouse Cedex**France
JOURNAL: Journal of Molecular Biology 260 (2):p178-195 1996
ISSN: 0022-2836
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: A growing number of small nucleolar RNAs (snoRNAs) are intron-encoded, contain the characteristic box C (UGAUGA) and box D (CUGA) motifs and exhibit long complementarities to conserved sequences in mature rRNAs. We have identified nine additional members of this family, U32 to U40. All but one are encoded in introns of ribosomal Protein genes in vertebrates: U32 to U35 in rpL13a, U36 in rpL7a and U38 to U40 in rpS8. By contrast, U37 is encoded in elongation factor 2 gene. Interestingly, U32 and U36 each contain two complementarities (one to 18 S and the other to 28 S rRNA). U32 to U40 are fibrillarin-associated, devoid of a 5'-trimethyl-cap and display an exclusively nucleolar localization. They are all metabolically **stable** and roughly as abundant as previously reported members of this family. Characterization of their homologs in distant species shows that their 10 to 14 nt long rRNA complementarities are conserved. A clue on the function of this

snORNA family is provided by the comparative analysis of the largely expanded collection of their conserved duplexes with rRNA. Not only does each duplex span at least one site of 2'-O-ribose methylation in the rRNA but the modification site is always at the same position in the duplex, paired to the fifth nucleotide upstream from a box D motif in the snoRNA. Consistent with the notion that each snoRNA of this family guides one particular methylation along the rRNA sequence, we have detected several pairs of snoRNAs with overlapping complementarities to rRNA tracts with vicinal sites of ribose methylations. In each case, the two overlapping complementarities are shifted from each other by a distance equal to the spacing between the methylated sites which are thus found at the same position within each of the mutually exclusive duplexes. Finally, we have also identified, within three previously known snoRNAs, novel antisense elements able to form a canonical duplex around ribose-methylated sites in rRNA, which further supports the conclusion that the duplex structure provides the 2'-O-methyltransferase with the appropriate site-specificity on the substrate.

1996

15/3,AB/4 (Item 2 from file: 5)
DIALOG(R) File 5:Biosis Previews(R)
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10267032 BIOSIS NO.: 199698721950
Synthesis of 2'-O-methylisocytidine phosphoramidite and
methylphosphonamidite synthons.
AUTHOR: Wang Daguang; Ts'o Paul O P(a)
AUTHOR ADDRESS: (a)Dep. Biochem., Sch. Hygiene Public Health, Johns Hopkins
Univ., 615 North Wolfe St., Baltimore, **USA
JOURNAL: Nucleosides & Nucleotides 15 (1-3):p387-397 1996
ISSN: 0732-8311
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The 2'-O-methylisocytidine phosphoramidite synthon 7 and
methylphosphonamidite synthon 8 are synthesized from 2'-O-methyluridine.
The N-2-(N',N'-dimethylformamidine) protected 2'-O-methylisocytidine is
stable to basic deamination and acidic depyrimidination. Synthon 7
and synthon 8 have been incorporated into oligomers via the automated
solid state procedure.

1996

15/3,AB/5 (Item 3 from file: 5)
DIALOG(R) File 5:Biosis Previews(R)
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10054727 BIOSIS NO.: 199598509645
6-Oxocytidine a novel protonated C-base analogue for **stable** triple
helix formation.
AUTHOR: Berressem Rainer; Engels Joachim W(a)
AUTHOR ADDRESS: (a)Johann Wolfgang Goethe-Univ. Frankfurt, Main**Germany
JOURNAL: Nucleic Acids Research 23 (17):p3465-3472 1995
ISSN: 0305-1048
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: 2'-O-Methyl-3'-O-phosphoramidite building blocks of 6-oxocytidine
6 and its 5-methyl derivative 7, respectively, were synthesized and

incorporated via phosphoramidite chemistry in 15 mer oligodeoxynucleotides (d(T-72T-7), S2; d(T-73T-7), S3) to obtain potential Py cndot Pu cndot Py triplex forming homopyrimidine strands. UV thermal denaturation studies and CD spectroscopy of 1:1 mixtures of these oligomers and a 21 mer target duplex (d(C-3A-7GA-7C-3)-d(G-3T-7CT-7G-3), D-1) with a complementary purine tract showed a nearly pH-independent (6.0-8.0) triple helix formation with melting temperatures of 21-190C and 18.5-17.5 degree C, respectively (buffer system: 50 mM sodium cacodylate, 100 mM NaCl, 20 mM MgCl₂). In contrast, with the corresponding 15mer deoxy-C-containing oligonucleotide (d(T-71T-7), S1) triplex formation was observed only below pH 6.6. Specificity for the recognition of Watson-Crick GCbase pairs was observed by pairing the modified C-bases of the 15mers with all other possible Watson-Crick-base compositions in the target duplex (d(C-3A-7XA-7C-3)-d(G-3T-7YT-7G-3), X = A,C,T; Y = T,G,A, D2-4). Additionally, the Watson-Crick-pairing of the modified oligomers S2 and S3 was studied.

1995

15/3,AB/6 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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09993090 BIOSIS NO.: 199598448008
Absorption, tissue distribution and in vivo stability in rats of a hybrid antisense oligonucleotide following oral administration.
AUTHOR: Agrawal Sudhir(a); Zhang Xueshu; Lu Zhihong; Zhao Hui; Tamburin Jeffrey M; Yan Jieming; Cai Hongying; Habus Robert B Vv Diasioan; Jiang Zhiwei; Iyer Radhakrishnan P; Yu Dong; Zhang Ruiwen
AUTHOR ADDRESS: (a)Hybridon, Inc., Worcester, MA 01605**USA
JOURNAL: Biochemical Pharmacology 50 (4):p571-576 1995
ISSN: 0006-2952
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: In vivo stability and oral bioavailability of an oligodeoxynucleotide phosphorothioate containing segments of 2'-O-methyloligonucleotide phosphorothioates at both the 3'- and 5'-ends (hybrid oligonucleotide) were studied. A 25-mer 35S-labeled hybrid oligonucleotide was administered to rats by gavage at a dose of 50 mg/kg body weight. HPLC analysis revealed that this hybrid oligonucleotide was stable in the gastrointestinal tract for up to 6hr following oral administration. Radioactivity associated with the hybrid oligonucleotide was detectable in portal venous plasma, systemic plasma, various tissues, and urine. Intact hybrid oligonucleotide was detected, by HPLC analysis, in portal venous plasma, systemic plasma, and various tissues. The majority of the radioactivity in urine was associated with degradative products with lower molecular weights, but the intact form was also detected. In summary, the hybrid oligonucleotide was absorbed intact through the gastrointestinal tract, indicating the possibility of oral administration of oligonucleotides, a finding that may be important in the development of antisense oligonucleotides as therapeutic agents.

1995

result from hydrophobic interactions between substituents in the minor groove. As the 2'-O-methyl groups are directed into the minor groove, it may be worthwhile to introduce tailor-made 2'-O-substituents into RNA; it might be possible to design groups that both stabilize the hybrid duplexes and carry a nuclease function, further improving the efficacy of these modified RNAs in antisense applications.

17/3,AB/2 (Item 2 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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06577518 90202792 PMID: 1690712
Kinetic analysis of *Escherichia coli* RNase H using DNA-RNA-DNA/DNA substrates.

Hogrefe H H; Hogrefe R I; Walder R Y; Walder J A
Department of Biochemistry, University of Iowa, Iowa City 52242.
Journal of biological chemistry (UNITED STATES) Apr 5 1990, 265
(10) p5561-6, ISSN 0021-9258 Journal Code: 2985121R
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The kinetic properties of *Escherichia coli* ribonuclease H (RNase H) were investigated using oligonucleotide substrates that consist of a short stretch of RNA, flanked on either side by DNA (DNA-RNA-DNA). In the presence of a complementary DNA strand, RNase H cleavage is restricted to the short ribonucleotide stretch of the DNA/RNA heteroduplex. The DNA-RNA-DNA substrate utilized for kinetic studies: (formula; see text) is cleaved at a single site (decreases) in the presence of a complementary DNA strand, to generate (dT)7-(rA)2-OH and p-(rA)2-(dT)9. Anion exchange high performance liquid chromatography was used to separate and quantitate the cleavage products. Under these conditions, RNase H-specific and nonspecific degradation products could be resolved. Kinetic parameters were measured under conditions of 100% hybrid formation (1.2-1.5 molar excess of complementary DNA, T much less than Tm). A linear double reciprocal plot was obtained, yielding a Km of 4.2 microM and a turnover number of 7.1 cleavages per s per RNase H monomer. The kinetic properties of substrate analogs containing varying lengths of RNA (n = 3-5) and 2'-O-methyl modifications were also investigated. Maximal turnover was observed with DNA-RNA-DNA substrates containing a minimum of four RNA residues. Kcat for the rA3 derivative was decreased by more than 100-fold. The Km appeared to decrease with the size of the internal RNA stretch (n = 3-5). No significant difference in turnover number of Km was observed when the flanking DNA was replaced with 2'-O-methyl RNA, suggesting that RNase H does not interact with this region of the heteroduplex.

17/3,AB/3 (Item 3 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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06151051 89166496 PMID: 3233211
Raman spectroscopic measurement of base stacking in solutions of adenosine, AMP, ATP, and oligoadenylylates.

Weaver J L; Williams R W
Department of Biochemistry, Uniformed Services University of the Health Sciences, Bethesda, Maryland 20814-4799.
Biochemistry (UNITED STATES) Dec 13 1988, 27 (25) p8899-903,
ISSN 0006-2960 Journal Code: 0370623

Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

Measurements of the colligative properties of nucleosides and their derivatives have shown that bases form transient aggregates in solution [Ts'o (1967) J. Am. Chem. Soc. 89, 3612-3622]. Aggregation of nucleotides cannot be measured by osmometry due to the presence of counterions. Sedimentation measurements are difficult to obtain and have been complicated by differences in pH [Ferguson et al. (1974) Biophys. Chem. 1, 325-337]. Raman studies of **oligonucleotides** have shown that the intensities due to base vibrational modes depend on the extent of base stacking, but this dependence has not been quantitated. We have measured this dependence by relating changes in the Raman spectra of nucleotides and nucleosides with previous measurements of colligative properties. Visible Raman spectra of ATP, AMP, and adenosine, taken over a range of concentrations from 1 to 1000 mM, show that the peak intensity ratio (I1305 + I1380)/I1340 varies linearly with the log of the concentration for all three bases. This concentration-dependent change correlates with published molal osmotic coefficient data for functionally similar bases with a correlation coefficient of 0.99. In contrast, UV resonance Raman spectra of the same bases show changes that vary linearly with concentration.

17/3,AB/4 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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10568824 BIOSIS NO.: 199699189969
Antisense 2'-O-methyloligonucleotides hybridized to RNA block a nuclear, ATP-dependent 3'-5' exonuclease.
AUTHOR: Dominski Zbigniew; Ferree Patrick; Kole Ryszard(a)
AUTHOR ADDRESS: (a)Univ. N.C., Lineberger Comprehensive Cancer Cent., CB 7295, Chapel Hill, NC 27599**USA
JOURNAL: Antisense & Nucleic Acid Drug Development 6 (1):p37-45 1996
ISSN: 1087-2906
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: RNA hybridized to 2'-O-methyloligonucleotides and incubated in nuclear extracts from HeLa cells is truncated, resulting in a distinct product terminated at the 5' end of the antisense **oligonucleotide**. The activity responsible for this effect is not RNase H but rather a novel exonuclease degrading RNA in the 3' to 5' direction. The enzyme requires ATP and Mg-2+ ions. Except for dATP, no other nucleoside triphosphate or nonhydrolyzable ATP analog supports the exonucleolytic activity. In spite of the nuclear origin and activity requirements similar to those required for pre-mRNA splicing, the exonuclease operates with equal efficiency on intron-containing and intronless RNAs, excluding the possibility that it is associated with the splicing machinery.

1996

17/3,AB/5 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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10107716 BIOSIS NO.: 199698562634
Structural basis for the RNA binding selectivity of **oligonucleotide** analogues containing alkylsulfide internucleoside linkages and 2'-substituted 3'-deoxyribonucleosides.
AUTHOR: Damha Masad J(a); Meng Bin; Wang Daguang; Yannopoulos Constantin G; Just George
AUTHOR ADDRESS: (a)Dep. Chem., McGill Univ., 801 Sherbrooke St. W., Montreal, PQ H3A 2K6**Canada
JOURNAL: Nucleic Acids Research 23 (19):p3967-3973 1995

ISSN: 0305-1048

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: In this report we describe the synthesis of oligonucleotides containing sulfide-linked dinucleoside units, namely rT-(2'OH)-sdT, rT-(2'OMe)-sdT, dT-srU-(2'OMe) and dT-(2'OMe)-srU-(2'OMe). We also describe the interactions of such oligomers with complementary DNA and RNA targets, and provide the structural basis for their remarkable RNA binding selectivity. In all cases, the T-m values of the S/P-chimera duplexes were lower than those of the corresponding unmodified duplexes. We attribute this to steric interactions between the 5' sulfur and the atoms of the nearby base/sugar residues. The 2'-substituents (i.e., 2'OH or 2'OMe) vicinal to the alkylsulfide internucleoside linkage significantly perturb the structure and stability of the duplexes formed with DNA, and more so than with RNA. The introduction of three rT-(2'OH)-sdT-p, (or rT-(2'OMe)-sdTD-p) units into an oligodeoxynucleotide sequence was sufficient to abolish binding to complementary DNA but not RNA. The same three substitutions with dT-srU-(2'OMe)-p, and dT-(2'OMe)-srU-(2'OMe)-p did not abolish binding to DNA but the resulting complexes had poor thermal stability. The RNA-binding 'selectivity' exhibited by these oligomers is attributed to the tendency of the 2'-substituted (branched) furanoses to adopt the C3'-endo pucker, a conformation that is inconsistent with the B-form structure of helical DNA. The preference of these sugars to exist often exclusively in the C3'-endo form is attributed to stereoelectronic effects, namely gauche and anomeric effects. Our findings support the hypothesis that nucleoside analogues puckered exclusively in the C3'-endo form may result in them being especially good binders of targeted mRNA (S.H. Kawai (1991), Ph.D. Thesis, McGill University; Kawasaki et al. (1993) J. Med. Chem. 36, 831-841).

1995

ds

| Set | Items | Description |
|-----|-------|----------------------------|
| S1 | 0 | PY<1997 AND "2'-O-METHYL?" |
| S2 | 0 | "2'-O-METHYL" |
| S3 | 773 | PY<1997 AND ?METHYL? |
| S4 | 0 | S3 AND "2" (w) "O" |
| S5 | 4869 | PY<1997 AND HAIRPIN? |
| S6 | 172 | S5 AND METHYL? |
| S7 | 18 | S6 AND DETECT? |
| S8 | 14 | RD (unique items) |
| S9 | 162 | 2'-O-METHYL? |
| S10 | 156 | RD (unique items) |
| S11 | 0 | S5 AND S10 |
| S12 | 51 | S10 AND PY<1997 |
| S13 | 0 | S12 AND HAIRPIN? |
| S14 | 4 | S12 AND LOOP? |
| S15 | 6 | S12 NOT S14 AND STABL? |
| S16 | 41 | S12 NOT S14 NOT S15 |
| S17 | 5 | S16 AND OLIGONUCLEOTIDE? |
| S18 | 21 | S5 AND PROBE AND STABL? |
| S19 | 12 | RD (unique items) |

? s s5 and antisens?

4869 S5

37483 ANTISENS?

S20 112 S5 AND ANTISENS?

? s s20 and stabl?

112 S20

353726 STABL?

S21 31 S20 AND STABL?

? rd

...completed examining records

S22 18 RD (unique items)

? t s22/3,ab/all

22/3,AB/1 (Item 1 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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10717739 97067123 PMID: 8910525

Mechanisms of inhibition of in vitro dimerization of HIV type I RNA by sense and antisense oligonucleotides.

Skripkin E; Paillart J C; Marquet R; Blumenfeld M; Ehresmann B; Ehresmann C

Unite Propre de Recherche 9002 du CNRS, Institut de Biologie Moleculaire et Cellulaire, 15 rue Rene Descartes, 67084 Strasbourg-cedex, France.
ehresmc@ibmc.u-strasbg.fr

Journal of biological chemistry (UNITED STATES) Nov 15 1996, 271
(46) p28812-7, ISSN 0021-9258 Journal Code: 2985121R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Retroviruses display a strong selective pressure to maintain the dimeric nature of their genomic RNAs, suggesting that dimerization is essential for viral replication. Recently, we identified the cis-element required for initiation of human immunodeficiency virus type I (HIV-I) RNA dimerization in vitro. The dimerization initiation site (DIS) is a hairpin structure containing a self-complementary sequence in the loop. We proposed that dimerization is initiated by a loop-loop kissing interaction involving the self-complementary sequence present in each monomer. We tested the ability of sense and antisense oligonucleotides targeted against the DIS to interfere with a preformed viral RNA dimer. Self-dimerization and inhibition properties of the tested oligonucleotides are dictated by the

nature of the loop. An RNA loop is absolutely required in the case of sense oligonucleotides, whereas the nature and the sequence of the stem is not important. They form reversible loop-loop interactions and act as competitive inhibitors. **Antisense** oligonucleotides are less efficient in self-dimerization and are more potent inhibitors than sense oligonucleotides. They are less sensitive to the nature of the loop than the **antisense** oligonucleotides. **Antisense hairpins** with either RNA or DNA stems are able to form highly **stable** and irreversible complexes with viral RNA, resulting from complete extension of base pairing initiated by loop-loop interaction.

22/3,AB/2 (Item 2 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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10423489 96229893 PMID: 8639652
Unambiguous structure characterization of a DNA-RNA triple helix by ¹⁵N- and ¹³C-filtered NOESY spectroscopy.

van Dongen M J; Heus H A; Wymenga S S; van der Marel G A; van Boom J H; Hilbers C W

NSR Centre for Molecular Structure, Design, and Synthesis, University of Nijmegen, The Netherlands.

Biochemistry (UNITED STATES) Feb 13 1996, 35 (6) p1733-9,
ISSN 0006-2960 Journal Code: 0370623

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

DNA.DNA*RNA triple helices of the pyrimidine.purine*pyrimidine motif (where . indicates Watson-Crick pairing and * indicates Hoogsteen pairing) appear to be very **stable**, which has important implications for the development of novel **antisense** strategies. Here we present the first structural NMR studies on such a system, composed of a DNA **hairpin** with a homopurine-homopyrimidine stem sequence and a single-stranded RNA oligonucleotide containing exclusively pyrimidine residues. In these investigations an unlabeled DNA **hairpin** and a uniformly ¹³C/¹⁵N-enriched RNA oligonucleotide were utilized in combination with X-edited ¹H NMR spectroscopy. Improved ¹⁵N (omega 2) filtered NOESY and ¹³C (omega .1) filtered NOESY are presented by which we were able to differentiate between intrastrand, i.e., DNA-DNA and RNA-RNA, and interstrand, i.e., DNA-RNA, NOE contacts. It is unambiguously established that the complex forms a right-handed triple helix, with the RNA strand situated in the major groove of the Watson-Crick stem of the **hairpin**. The interaction is stabilized by the formation of Hoogsteen-type base pairs between the RNA strand and the purine strand of the DNA. These strands run parallel to each other. The characterization of the DNA-RNA triple helix structure described here shows that this type of experiment forms a valuable instrument in the structure determination of bimolecular systems of nucleic acids.

22/3,AB/3 (Item 3 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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10311170 96113282 PMID: 8785472

4'-Thio-RNA: synthesis of mixed base 4'-thio-oligoribonucleotides, nuclease resistance, and base pairing properties with complementary single and double strand.

Leydier C; Bellon L; Barascut J L; Morvan F; Rayner B; Imbach J L
Laboratoire de Chimie Bio-Organique, URA 488, CNRS, Universite de Montpellier II, France.

Antisense research and development (UNITED STATES) Fall 1995, 5

(3) p167-74, ISSN 1050-5261 Journal Code: 9110698

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

4'-Thio-beta-D-oligonucleotides (12 mer and 16 mer) containing a mixed base sequence were synthesized via the phosphoramidite solid support approach. These RNA analogs showed very good nuclease resistance as compared with wild-type RNA. Furthermore, 4'-thio-beta-D-oligonucleotides were shown to hybridize with a complementary DNA or RNA strand to form a duplex or with a DNA hairpin to form a triple helix. 4'-Thio-RNA binds more tightly to its complementary RNA strand than to its complementary DNA strand. A 4'-thio-RNA:RNA duplex is as stable as a 2'-O-methyl-RNA:RNA duplex. 4'-Thio-RNA, however, forms a 4'-thio-RNA:DNA:DNA triplex with a stability similar to the corresponding triplex with all wild-type DNA.

22/3,AB/4 (Item 4 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08703554 95392159 PMID: 7545040

Exceptionally **stable** nucleic acid hairpins.

Varani G

MRC Laboratory of Molecular Biology, Cambridge, United Kingdom.

Annual review of biophysics and biomolecular structure (UNITED STATES)

1995, 24 p379-404, ISSN 1056-8700 Journal Code: 9211097

Document type: Journal Article; Review; Review, Academic

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Hairpins represent the dominant secondary structure element in RNA. Certain sequences are found with exceptional frequency in many RNAs and are characterized by exceptionally high thermodynamic stability. **Stable** RNA hairpins define nucleation sites for folding, determine tertiary interactions in RNA enzymes, protect mRNAs from degradation, and are recognized by RNA-binding proteins. The structures of several **stable** DNA and RNA hairpins have revealed networks of stabilizing interactions within the hairpin loop: non-Watson-Crick base pairs and base-phosphate and base-sugar contacts. The unusual stability of these structural elements can be used to stabilize RNA and DNA structures and to protect **antisense** oligonucleotides and mRNAs against exonucleolytic degradation.

22/3,AB/5 (Item 5 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08499684 95187959 PMID: 7882142

In vitro selection of **antisense** oligonucleotides targeted to a hairpin structure.

Mishra R K; Toulme J J

Laboratoire de Biophysique Moleculaire, INSERM U. 386, Universite Bordeaux-II, France.

Comptes rendus de l'Academie des sciences. Serie III, Sciences de la vie (FRANCE) Nov 1994, 317 (11) p977-82, ISSN 0764-4469

Journal Code: 8503078

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Antisense oligonucleotides are widely used to selectively prevent

pre-RNA splicing, mRNA translation or cDNA synthesis from a retroviral RNA template. However, intramolecular folding of the RNA chain can sequester the target sequence into a stable structure. Consequently, the antisense effect can be greatly reduced or even abolished. Hydrogen donor and acceptor sites are still available on nucleic acid bases involved in secondary structures. However, the rational design of antisense sequences able to recognize the three dimensional array of these sites is not available. We used an in vitro selection procedure to fish out aptastructs, i.e., oligomers able ("apte") to bind to a structure. A population of randomly synthesized oligonucleotides was mixed with the structure of interest and oligodeoxynucleotide sequences bound to the target were selected and amplified. The selection involves the destruction of the unbound candidates by a restriction enzyme. This procedure can be used both for RNA and DNA target structures and does not require the purification of the bound oligonucleotides at each cycle of selection. Several cycles of selection-amplification, followed by cloning and sequencing, allowed us to identify three oligonucleotides able to form a complex with a DNA hairpin. Due to the sequence of the selected candidates, these aptastruct-hairpin complexes involve very likely non-canonical interactions between the two partners.

22/3,AB/6 (Item 6 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08464044 95152260 PMID: 7849485

Improved anti-herpes simplex virus type 1 activity of a phosphodiester antisense oligonucleotide containing a 3'-terminal hairpin-like structure.

Poddevin B; Meguenni S; Elias I; Vasseur M; Blumenfeld M

Department of Therapeutical Research, GENSET, Paris, France.

Antisense research and development (UNITED STATES) Fall 1994, 4

(3) p147-54, ISSN 1050-5261 Journal Code: 9110698

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

We synthesized a series of 20-mer antisense phosphodiester oligonucleotides constituting of a 5'-dodecameric sequence, complementary to the acceptor splice junction of herpes simplex virus type 1 (HSV-1) pre-mRNAs IE4 and IE5, flanked in 3' by octameric sequences adopting hairpin-like structures of different stabilities. The presence of the minihairpins in 3' protected the 20-mer phosphodiester oligonucleotides against serum nuclease degradation, this protection being well correlated to the reported melting temperatures of the minihairpins, and to the gel mobilities of the 20-mer oligonucleotides. While no protection was observed using a linear 8-mer, the addition in 3' of the most stable minihairpin--H8--increased more than eightfold the nuclease resistance of the linear antisense dodecamer. We analyzed the effect of such a protection on the anti-HSV-1 antisense activities of the oligonucleotides. When bearing H8 in 3', the antisense dodecamer was 10 times more active than in the absence of 3'-flanking sequence, while a linear 20-mer control containing the antisense sequence was only 3 times more active. This work provides the basis for a further rational design of phosphodiester antisense oligonucleotides, taking advantage of the specific properties conferred by their conformations.

22/3,AB/7 (Item 7 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08430926 95119015 PMID: 7819224

Recognition and cleavage of single-stranded DNA containing **hairpin** structures by oligonucleotides forming both Watson-Crick and Hoogsteen hydrogen bonds.

Francois J C; Helene C
Laboratoire de Biophysique, Museum National d'Histoire Naturelle, INSERM
U 201, CNRS UA 481, Paris, France.

Biochemistry (UNITED STATES) Jan 10 1995, 34 (1) p65-72,
ISSN 0006-2960 Journal Code: 0370623

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

A new approach is described to design **antisense** oligonucleotides targeted against single-stranded nucleic acids containing **hairpin** structures by use of both Watson-Crick and Hoogsteen hydrogen bond interactions for recognition. The oligonucleotide has two different domains, one allowing double helix formation involving Watson-Crick base pairs and the other one forming a triple helix involving Hoogsteen-type base triplets in the major groove of a **hairpin** stem. Spectroscopic and gel retardation experiments provided evidence for such Watson-Crick/Hoogsteen (WC/H) recognition of **hairpin** structures in single-stranded DNA. An **antisense** oligonucleotide designed to form only Watson-Crick base pairs was unable to disrupt the **stable** stem structure of the target under conditions where the oligonucleotide designed with the Watson-Crick/Hoogsteen interactions could bind efficiently to the **hairpin** -containing target. The addition of one nucleotide to the oligonucleotide at the junction between the double helix and triple helix regions in WC/H complexes had an effect on stability which was dependent on the relative orientation of the Watson-Crick and Hoogsteen domains in the target. An oligodeoxynucleotide-phenanthroline conjugate targeted against such a **hairpin**-containing DNA fragment induced specific cleavage in the double-stranded stem. This WC/H approach may be useful in designing artificial regulators of gene expression.

22/3,AB/8 (Item 8 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08428230 95116319 PMID: 7816620

Evolutionarily conserved elements in the 5' untranslated region of beta globin mRNA mediate site-specific priming of a unique **hairpin** structure during cDNA synthesis.

Volloch V Z; Schweitzer B; Rits S
Boston Biomedical Research Institute, MA 02114.
Nucleic acids research (ENGLAND) Dec 11 1994, 22 (24) p5302-9,
ISSN 0305-1048 Journal Code: 0411011

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Generation of double-stranded cDNA during reverse transcription of a variety of mRNA molecules is well known to involve the formation of covalently linked **antisense** and sense strands in a **hairpin** configuration. In the present study we have examined the sequence of molecular events which occurs during cDNA synthesis from mouse beta globin mRNA, in particular the self-priming event that initiates synthesis of sense-strand DNA. Upon completion of reverse transcription of globin mRNA and the removal of RNA template by RNase H activity associated with reverse transcriptase, the 3' end of cDNA snaps back to form a **stable** double-stranded structure, which is extended by reverse transcriptase to generate the sense DNA strand. Surprisingly, the fourteen 3' terminal nucleotides of the beta globin **antisense** DNA strand (cDNA) have strong complementarity with an internal segment of the same molecule

corresponding to a portion of the 5'-untranslated region of the mRNA located just upstream of the translation start site. Efficient second strand cDNA synthesis appears to require the occurrence within the cDNA molecule of these two complementary elements, one of which must be 3'-terminal. A second surprising feature is that the strong complementarity between the terminal and the internal portions of the molecule exists in the **antisense** DNA and not in the sense mRNA strand. This is because A:C mismatches on the sense strand correspond to relatively **stable** T:G base pairs on the **antisense** strand. Such an extended region of complementarity within the segment of cDNA corresponding to the short 5' untranslated region of beta globin mRNA is unlikely to occur purely by chance, suggesting some underlying function. In this regard it is of interest that cDNAs of adult beta globin mRNAs from other mammalian species show a very similar arrangement of complementary elements, and that complementarity is heavily conserved, even when there are substitutions in nucleotide sequence.

22/3,AB/9 (Item 9 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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08377670 95065643 PMID: 7975207
Activity and cleavage site specificity of an anti-HIV-1 **hairpin** ribozyme in human T cells.
Yamada O; Kraus G; Leavitt M C; Yu M; Wong-Staal F
Department of Medicine and Biology, University of California, San Diego
92093-0665.
Virology (UNITED STATES) Nov 15 1994, 205 (1) p121-6, ISSN
0042-6822 Journal Code: 0110674
Contract/Grant No.: AI 31378; AI; NIAID
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed
Human CD4+ T cells (Molt-4) were transduced with retroviral vectors containing a **hairpin** ribozyme which targets the rev/env coding region of HIV-1 RNA (HXB2: 8629-8644). This target sequence is conserved among many HIV-1 clones, including the prototype virus HXB2, but the infectious clone SF2 contains a single nucleotide substitution at the cleavage site (from N*GUC to N*UUC). Cells **stably** expressing the ribozyme or its disabled counterpart were challenged with HXB2 or SF2 and the amount of p24 antigen produced was monitored. While this ribozyme was effective in inhibiting the replication of HXB2 in Molt 4 cells, it showed only marginal inhibitory effect on SF2 replication. The same level of virus production was observed with cells transduced by the disabled ribozyme, which functions essentially as an **antisense** molecule. Expression of the ribozyme was comparable in HXB2- or SF2-infected cells as detected by reverse transcription-polymerase chain reaction. These data provide *in vivo* evidence that the antiviral activity of the **hairpin** ribozyme is strictly dependent on the presence of the cleavage site in the target RNA and supports the conclusion that the ribozyme acts as catalytic RNA rather than as **antisense** RNA *in vivo*.

22/3,AB/10 (Item 10 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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08302530 94368806 PMID: 8086420
Specific inhibition of expression of a human collagen gene (COL1A1) with modified **antisense** oligonucleotides. The most effective target sites are clustered in double-stranded regions of the predicted secondary structure for the mRNA.

Laptev A V; Lu Z; Colige A; Prockop D J
Department of Biochemistry and Molecular Biology, Jefferson Institute of
Molecular Medicine, Jefferson Medical College, Thomas Jefferson University,
Philadelphia, Pennsylvania 19107.
Biochemistry (UNITED STATES) Sep 13 1994, 33 (36) p11033-9,
ISSN 0006-2960 Journal Code: 0370623

Contract/Grant No.: AR-38188; AR; NIAMS; AR-39740; AR; NIAMS
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

A series of antisense oligonucleotides (ASOs) were synthesized and tested to define the best target sites within an RNA transcript of collagen for effective inhibition of expression. The test system consisted of mouse NIH 3T3 fibroblasts that were stably transfected with a human minigene for procollagen I so that the cells simultaneously synthesized full-length mouse pro alpha 1 (I) chains and internally deleted human pro alpha 1 (I) chains. The sequences of the transcripts from both genes were compared, and a series of 28 ASOs were designed to target sites in which there were at least two base differences within a 20-nucleotide sequence between the human and mouse transcripts. Six of the ASOs specifically decreased the levels of pro alpha 1 (I) chain synthesized from the human gene without a decrease in the levels of pro alpha 1 (I) chains from the mouse endogenous gene. The most effective ASOs reduced the intracellular levels of human pro alpha 1 (I) chains relative to the mouse pro alpha 1 (I) chains to 37-67% of the control values. Combined addition of two effective ASOs or a second administration of the same effective ASO did not produce any additive effect. The results did not support previous suggestions that the best target sites for ASOs were sequences around initiation codons for translation, at intron-exon boundaries, or in single-stranded loops in hairpin structures. Also, the results did not support previous suggestions that the most effective ASOs are those with the highest affinities for their target sequences. Instead, the most consistent pattern in the data was that the most effective ASOs were those targeted to sequences that were predicted to form clustered double-stranded structures in RNA transcripts.

22/3,AB/11 (Item 11 from file: 155)
DIALOG(R) File 155: MEDLINE(R)
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08293565 94359804 PMID: 8078767
Pyrimidine phosphorothioate oligonucleotides form triple-stranded helices and promote transcription inhibition.

Xodo L; Alunni-Fabbroni M; Manzini G; Quadrifoglio F
Department of Biochemistry, Biophysics and Macromolecular Chemistry,
University of Trieste, Italy.
Nucleic acids research (ENGLAND) Aug 25 1994, 22 (16) p3322-30
ISSN 0305-1048 Journal Code: 0411011
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: Completed

The ability of phosphorothioate (POS) oligonucleotides to recognise and bind to homopurine-homopyrimidine DNA double-stranded sites via triple helix formation has been investigated. It has been found that the homologous pyrimidine POS sequences Y11-Si (i = 0, 1, 2, 3, 4, 10), which have been obtained by an increasing sulphur substitution in the sugar-phosphate backbone of d(CTTCCTCCTCT) (Y11), and the target hairpin duplex d(GAAGGAGGAGA-T4-TCTCCTCCTTC) (h26) can form stable triple helices, as indicated by PAGE, CD and UV melting experiments. The thermal stability of the triple helices depends on the number of POS linkages in the third Y11 strand, varying from 48 degrees C (Y11, with only phosphate groups,

PO2) to 31 degrees C (Y11-S10 containing exclusively thioate groups). On average, a Tm depression of about 2 degrees C per POS linkage introduced in Y11 was observed. CD data indicate that the sulphurization of the third strand results in minimal changes of triple-stranded structures. The energetics of the triplex-to-hairpin plus single-strand transition has been determined by van't Hoff analyses of the melting curves. In free energy terms, the POS triplexes h26.Y11-Si are less stable than the normal PO2 h26.Y11 triplex by values between 2.7 and 5.4 kcal/mol, depending on the number of POS linkages contained in the third strand. Phosphorothioate oligonucleotides being resistant towards several nucleases offer an interesting choice as gene blockers in antisense strategy. Thus, their ability to inhibit transcription via triple helix formation has been examined in vitro. We found that triplex-forming POS oligonucleotides of 20 bases in length (with a cytosine contents of 45%), containing either 10% or 26% thioate groups, strongly repress the transcription activity of the bacteriophage T7 RNA polymerase at pH 6.9, when used in excess compared to the target (mol oligo/mol template = 125). The here reported data are useful for designing phosphorothioate oligonucleotides targeted to genomic DNA in antigene strategy.

22/3,AB/12 (Item 12 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08121294 94187059 PMID: 8139004

Nucleotide sequence and structural determinants of specific binding of coat protein or coat protein peptides to the 3' untranslated region of alfalfa mosaic virus RNA 4.

Houser-Scott F; Baer M L; Liem K F; Cai J M; Gehrke L
Division of Health Sciences and Technology, Massachusetts Institute of Technology, Cambridge 02139.

Journal of virology (UNITED STATES) Apr 1994, 68 (4) p2194-205

, ISSN 0022-538X Journal Code: 0113724

Contract/Grant No.: GM42504; GM; NIGMS

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

The specific binding of alfalfa mosaic virus coat protein to viral RNA requires determinants in the 3' untranslated region (UTR). Coat protein and peptide binding sites in the 3' UTR of alfalfa mosaic virus RNA 4 have been analyzed by hydroxyl radical footprinting, deletion mapping, and site-directed mutagenesis experiments. The 3' UTR has several stable hairpins that are flanked by single-stranded (A/U)UGC sequences.

Hydroxyl radical footprinting data show that five sites in the 3' UTR of alfalfa mosaic virus RNA 4 are protected by coat protein, and four of the five protected regions contain AUGC or UUGC. Electrophoretic mobility band shift results suggest four coat protein binding sites in the 3' UTR. A 3'-terminal 39-nucleotide RNA fragment containing four AUGC repeats bound coat protein and coat protein peptides with high affinity; however, coat protein bound poorly to antisense 3' UTR transcripts and poly(AUGC)10. Site-directed mutagenesis of AUGC865-868 resulted in a loss of coat protein binding and peptide binding by the RNA fragment. Alignment of alfalfa mosaic RNA sequences with those from several closely related ilarviruses demonstrates that AUGC865-868 is perfectly conserved; moreover, the RNAs are predicted to form similar 3'-terminal secondary structures. The data strongly suggest that alfalfa mosaic virus coat protein and ilarvirus coat proteins recognize invariant AUGC sequences in the context of conserved structural elements.

22/3,AB/13 (Item 13 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

07189881 92052231 PMID: 1946436

Efficient trans cleavage and a common structural motif for the ribozymes of the human hepatitis delta agent.

Branch A D; Robertson H D

Center for Studies of the Biological Correlates of Addiction, Rockefeller University, New York, NY 10021.

Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) Nov 15 1991, 88 (22) p10163-7, ISSN 0027-8424 Journal Code: 7505876

Contract/Grant No.: DA-5130; DA; NIDA

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Cis-active ribozymes are potential therapeutic agents; however, to be used in this capacity, they must first be converted to trans-active ribozymes, a process facilitated by analysis of their structures. We present evidence that the genomic and antigenomic ribozymes of the human delta hepatitis agent share a structural ("axehead") motif that has conserved sequence elements and a **stable hairpin**. Guided by the features of the axehead, we divided each of the delta ribozymes into two subdomains, which we synthesized as separate RNA transcripts to give an enzyme and substrate for each ribozyme. Incubation of a substrate subdomain with its matching enzyme resulted in efficient and accurate trans cleavage. This work forms the basis for kinetic studies and for adapting the delta ribozymes for cleavage of selected target RNAs.

22/3,AB/16 (Item 16 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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07038587 91279451 PMID: 2057355

Use of electrophoretic mobility to determine the secondary structure of a small **antisense** RNA.

Jacques J P; Susskind M M

Department of Biological Sciences, University of Southern California, Los Angeles 90089-1340.

Nucleic acids research (ENGLAND) Jun 11 1991, 19 (11) p2971-7, ISSN 0305-1048 Journal Code: 0411011

Contract/Grant No.: GM3681; GM; NIGMS

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Natural **antisense** RNAs have stem-loop (**hairpin**) secondary structures that are important for their function. The **sar antisense** RNA of phage P22 is unusual: the 3' half of the molecule forms an extensive stem-loop, but potential structures for the 5' half are not predicted to be thermodynamically **stable**. We devised a novel method to determine the secondary structure of **sar** RNA by examining the electrophoretic mobility on non-denaturing gels of numerous **sar** mutants. The results show that the wild-type RNA forms a 5' stem-loop that enhances electrophoretic mobility. All mutations that disrupt the stem of this **hairpin** decrease mobility of the RNA. In contrast, mutations that change the sequence of the stem without disrupting it (e.g. change G.U to A.U) do not affect mobility. Nearly all mutations in single-stranded regions of the structure also have no effect on mobility. Confirmation of the proposed 5' stem-loop was obtained by constructing and analyzing compensatory double mutants. Combinations of mutations that restore a base-pair of the stem also restore mobility. The genetic phenotypes of **sar** mutants confirm that the proposed secondary structure is correct and is essential for optimal activity of the **antisense** RNA *in vivo*.

gives a T_m close to that found for the genomic RNA isolated from the virions. Thermodynamic parameters derived from denaturation curves of dimers also suggest that dimerization involves short sequences. The conformation of RNA fragments containing the 5' untranslated region and the 5' Gag-coding region has been probed by the use of a large variety of chemical structure probes. Secondary structural models have been proposed for RNA from HIV1 and MoMuLV. In both systems, RNA can be folded in structural domains containing the different functional sites.

Dimerization induces structural rearrangements. In HIV1, the TAR and PBS are **stable hairpin** structures, while the dimer linkage structure and the Gag translational initiation domain undergo structural rearrangements. These structural rearrangements are discussed with respect to the proposed mechanism of dimerization and their functional implications. In MoMuLV, structural rearrangements are also evidenced in the Psi and translational initiation domains. The PBS is involved in a **stable structure** within the dimer, whereas in the monomer form this region can adopt alternative conformations.

1992

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| Set | Items | Description |
|-----------------------------|--------|--------------------------------|
| S1 | 0 | PY<1997 AND "2'-O-METHYL?" |
| S2 | 0 | "2'-O-METHYL" |
| S3 | 773 | PY<1997 AND ?METHYL? |
| S4 | 0 | S3 AND "2" (W) "O" |
| S5 | 4869 | PY<1997 AND HAIRPIN? |
| S6 | 172 | S5 AND METHYL? |
| S7 | 18 | S6 AND DETECT? |
| S8 | 14 | RD (unique items) |
| S9 | 162 | 2'-O-METHYL? |
| S10 | 156 | RD (unique items) |
| S11 | 0 | S5 AND S10 |
| S12 | 51 | S10 AND PY<1997 |
| S13 | 0 | S12 AND HAIRPIN? |
| S14 | 4 | S12 AND LOOP? |
| S15 | 6 | S12 NOT S14 AND STABL? |
| S16 | 41 | S12 NOT S14 NOT S15 |
| S17 | 5 | S16 AND OLIGONUCLEOTIDE? |
| ? s s5 and probe and stabl? | | |
| | 4869 | S5 |
| | 171317 | PROBE |
| | 353726 | STABL? |
| S18 | 21 | S5 AND PROBE AND STABL? |
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| | S19 | 12 RD (unique items) |
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19/3,AB/1 (Item 1 from file: 155)
DIALOG(R)File 155: MEDLINE(R)
(c) format only 2003 The Dialog Corp. All rts. reserv.

10811380 97101110 PMID: 8945637
Human kinesin light (beta) chain gene: DNA sequence and functional characterization of its promoter and first exon.

Chernajovsky Y; Brown A; Clark J
Kennedy Institute of Rheumatology, Molecular Biology Laboratory, Hammersmith, London, UK.

DNA and cell biology (UNITED STATES) Nov 1996, 15 (11) p965-74
, ISSN 1044-5498 Journal Code: 9004522

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Kinesins are tubulin molecular motors whose function is to transport organelles within cells. Very little is known about the regulation of expression of these proteins. We have characterized the gene product of one differentially spliced mRNA of the human light chain kinesin and cloned its promoter region. A full-length kinesin cDNA was translated in vitro in a cell-free system, producing a 70-kDa protein. Using this cDNA as a probe, we isolated and sequenced the promoter, first exon, and part of the first intron of this gene from a genomic lambda EMBL3 human placental DNA library. The whole gene spans more than 90 kb. The beta kinesin promoter region confers only constitutive transcription to the bacterial chloramphenicol acetyltransferase (CAT) reporter gene. In permanently transfected human HeLa and NB100 neuroblastoma cells, a reporter gene containing the promoter and part of the first exon of beta kinesin was 75-fold more active than the HSV-tk promoter. The first exon contains the 5'-untranslated sequence capable of forming a **stable double-hairpin** loop, which functions as a translational enhancer. Its deletion decreases the efficiency of in vitro translation of beta kinesin mRNA and confers increased translation to a CAT reporter gene.

19/3,AB/2 (Item 2 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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10621138 96438775 PMID: 8841120

Characterization of the metal ion binding helix-hairpin-helix motifs in human DNA polymerase beta by X-ray structural analysis.

Pelletier H; Sawaya M R

Department of Chemistry and Biochemistry, University of California, San Diego, La Jolla 92093-0506, USA.

Biochemistry (UNITED STATES) Oct 1 1996, 35 (39) p12778-87,

ISSN 0006-2960 Journal Code: 0370623

Contract/Grant No.: GM52860; GM; NIGMS

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

X-ray crystallographic studies have shown that DNA binding by human polymerase beta (pol beta) occurs primarily through two structurally and sequentially homologous helix-hairpin-helix (HhH) motifs, one in the fingers subdomain and the other in the 8-kDa domain [Pelletier, H., Sawaya, M. R., Wolfle, W., Wilson, S. H., & Kraut, J. (1996a) Biochemistry 35, 12742-12761]. In that DNA binding by each HhH motif is facilitated by a metal ion, we set out to determine the identity of the metal ion that most likely binds to the HhH motif in vivo. Crystal soaking experiments were performed on human pol beta-DNA cocrystals with Mg²⁺, Ca²⁺, Na⁺, and K⁺, the four most prevalent metal ions in the cell, and in each case a data set was collected and the resulting structure was refined. Under the conditions tested, the HhH motifs of pol beta have an affinity for these biologically prevalent metal ions in the order Mg²⁺ < Ca²⁺ < Na⁺ < K⁺, with K⁺ displaying the strongest binding. Crystals soaked in the presence of Tl⁺, a commonly used spectroscopic probe for K⁺, were too X-ray-sensitive to establish the binding behavior of Tl⁺, but soaking experiments with Ba²⁺ and Cs⁺ resulted in relatively stable crystals that gave evidence of metal ion binding in both HhH motifs, confirming that larger monovalent and divalent metal ions are capable of binding to the HhH metal sites. Although Mn²⁺, which has been categorized as a potent polymerase mutagen, binds to the HhH motifs with a greater affinity than Mg²⁺, Mn²⁺ does not bind to the HhH motifs in the presence of equimolar concentrations of Na⁺. These results suggest that in vivo, where Mn²⁺ is present only in trace amounts, Mn²⁺ probably does not have a large effect on DNA binding and may instead manifest a mutagenic effect on pol beta primarily by distorting nucleotide binding or by directly affecting the catalytic step [Pelletier, H., Sawaya, M. R., Wolfle, W., Wilson, S. H., & Kraut, J. (1996b) Biochemistry 35, 12762-12777]. Crystal soaking experiments with 31-kDa apoenzyme crystals show that, in the absence of DNA, the HhH motif in the fingers subdomain binds metal ions with either much lower occupancy or not at all, indicating that metal ion binding is dependent on the presence of the DNA substrate.

19/3,AB/3 (Item 3 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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10383583 96188872 PMID: 8614641

Folding of the HDV antigenomic ribozyme pseudoknot structure deduced from long-range photocrosslinks.

Bravo C; Lescure F; Laugaa P; Fourrey J L; Favre A

Laboratoire de Photobiologie Moleculaire, Institute Jacque Monod, CNRS, Universite Paris, France.

Nucleic acids research (ENGLAND) Apr 1 1996, 24 (7) p1351-9,
ISSN 0305-1048 Journal Code: 0411011

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

A trans-acting system has been designed in order to explore the three-dimensional structure of the anti-genomic HDV ribozyme. In this system, the substrate (SANT) is associated by base-pairing to the catalytic RNA (RzANT) forming helix H1. RzANT is able to cleave specifically the RNA substrate as well as a deoxysubstrate analogue containing a single ribocytidine at the cleavage site (position -1). This demonstrates that such deoxysubstrate analogues are valuable tools for structural studies of this ribozyme domain. They form however weak complexes with RzANT which is due in part to their ability to fold as **stable hairpins** unlike the RNA substrate. Using a set of full deoxy or of mixed deoxy-ribo substrate analogues site-specific substituted with the photoaffinity probe deoxy-4-thiouridine, ds4U, at a defined position, we were able to determine a number of long range contacts between the substrate and the ribozyme core. In particular, crosslinks between substrate position -1 and position -2 with residues C15, G19 and C67, thought to be involved in the ribozyme catalytic site, were detected. A three dimensional model of the antigenomic ribozyme system, derived from the structure proposed by Tanner et al. [Current Biol (1994) 4, 488-498] for the genomic system was constructed. Apart from residue deletion or insertion, only minor accommodations were needed to account for all photocrosslinks but one which is attributed to an alternative hybridization of the substrate with the ribozyme. This study therefore further supports the structure proposed by Tanner et al. for the pseudoknot model.

19/3,AB/4 (Item 4 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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08367774 95055746 PMID: 7966321

Structure-mapping of the **hairpin** ribozyme. Magnesium-dependent folding and evidence for tertiary interactions within the ribozyme-substrate complex.

Butcher S E; Burke J M

Department of Microbiology and Molecular Genetics, Markey Center for Molecular Genetics, University of Vermont, Burlington 05405.

Journal of molecular biology (ENGLAND) Nov 18 1994, 244 (1)
p52-63, ISSN 0022-2836 Journal Code: 2985088R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

We have used chemical modification analysis to **probe** the solution structure of the **hairpin** ribozyme. The modifying reagents dimethylsulfate, 1-cyclohexyl-N'-(2-(N-methylmorpholino) ethyl-carbodiimide-p-toluenesulfonate, kethoxal, diethylpyrocarbonate and (2,12-dimethyl-3,7,11,17-tetraazabicyclo [11.3.1]heptadeca-1(17),2,11,13,15-pentaenato) nickel(II) perchlorate were used to **probe** functional groups that participate in Watson-Crick and non-canonical base-pairs. Our results confirm the existence of four short helices (3 to 6 bp) within the ribozyme-substrate complex, and demonstrate that one intramolecular helix (helix 4) is comprised of three base-pairs rather than the previously suggested five. In the absence of magnesium, the ribozyme is observed to fold into its secondary structure. Upon addition of magnesium, a striking difference in chemical modification is observed, particularly at sites within the ribozyme's large internal loop (loop B) that are essential for catalytic function (bases 21 to 26). Moreover, magnesium-dependent folding clearly destabilizes an A-U base-pair in a region where a proposed bend is required to juxtapose the catalytically essential loops A and B. Upon addition of substrate, no changes are observed in the structure of helix 3,

loop B or helix 4. However, strong protection of bases in the substrate-binding domain is observed, including those located across internal loop A. The modification data are consistent with the formation of a previously proposed tertiary structure motif within loop B that includes non-canonical G-A, A-U and A-A base-pairs, and that is identical with those identified by NMR analysis of loop E of 5 S rRNA and the sarcin/ricin loop of 28 S rRNA. Our results indicate that the **hairpin** ribozyme adopts a **stable** magnesium-dependent tertiary structure to which the substrate binds without inducing major conformational changes, and that substrate recognition is likely to involve non-canonical base-pairs between the ribozyme and substrate sequences adjacent to the cleavage site.

19/3,AB/5 (Item 5 from file: 155)
DIALOG(R)File 155: MEDLINE(R)
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08330688 95018658 PMID: 7933128

Sequence requirements for **stable** binding and function of Rep68 on the adeno-associated virus type 2 inverted terminal repeats.

Chiaroni J A; Wiener S M; Owens R A; Kyostio S R; Kotin R M; Safer B
Molecular Hematology Branch, National Heart, Lung, and Blood Institute,
Bethesda, MD 20892-1654.

Journal of virology (UNITED STATES) Nov 1994, 68 (11) p7448-57 .
, ISSN 0022-538X Journal Code: 0113724

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Replication of the palindromic inverted terminal repeats (ITRs) of adeno-associated virus type 2 requires several functions of the viral nonstructural Rep proteins. These include binding to the ITR, nicking of the double-stranded replication intermediate at the terminal resolution site (trs), and then strand displacement and synthesis from the nick. This report demonstrates the ability of both recombinant fusion maltose-binding protein (MBP)-Rep68 delta produced in Escherichia coli and wild-type (wt) Rep68 to bind to a linear truncated form of the ITR, delta 57 ITR, with similar affinity as to the wt **hairpin** ITR. A dissociation constant for MBP-Rep68 delta of approximately 8×10^{-10} M was determined for the wt ITR and delta 57 ITR probes. Truncation of delta 57 ITR to generate delta 28 ITR, which retains the GCTC repeat motif but not the trs, bound at least 10 times less efficiently than delta 57 ITR. Extension of delta 28 ITR with nonspecific sequence restored the ability of MBP-Rep68 delta to bind to delta 28 ITR. Thus, high-affinity binding would appear to require stabilization by flanking sequence as well as the intact GCTC repeat motif. Cleavage of the delta 57 ITR **probe** with DdeI, which truncates the flanking sequence and was previously shown to inhibit binding by Rep68, also inhibited the binding of MBP-Rep68 delta. The requirements for **stable** binding were further defined with a series of oligonucleotide probes which spanned the region protected by MBP-Rep78 in DNase I footprinting. The binding activity of either MBP-Rep68 delta or wt Rep68 to **hairpin** ITR or delta 57 ITR was indistinguishable. However, the binding activity of MBP-Rep68 delta to DNA does not appear to correlate with trs endonuclease activity. The nicking and covalent linkage of MBP-Rep68 delta to the nonhairpin delta 57 ITR was approximately 100-fold less efficient than its linkage to a **hairpin** -containing ITR. Therefore, although the **hairpin** portion of the ITR does not appear to play a role in recognition and stabilization of MBP-Rep68 delta binding, its presence does affect the trs cleavage activity of the protein.

19/3,AB/6 (Item 6 from file: 155)
DIALOG(R)File 155: MEDLINE(R)
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08195505 94261427 PMID: 8202361

Probing the hammerhead ribozyme structure with ribonucleases.

Hodgson R A; Shirley N J; Symons R H

Department of Plant Science, Waite Institute, University of Adelaide, Australia.

Nucleic acids research (ENGLAND) May 11 1994, 22 (9) p1620-5,
ISSN 0305-1048 Journal Code: 0411011

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Susceptibility to RNase digestion has been used to **probe** the conformation of the hammerhead ribozyme structure prepared from chemically synthesised RNAs. Less than about 1.5% of the total sample was digested to obtain a profile of RNase digestion sites. The observed digestion profiles confirmed the predicted base-paired secondary structure for the hammerhead. Digestion profiles of both *cis* and *trans* hammerhead structures were nearly identical which indicated that the structural interactions leading to self-cleavage were similar for both systems. Furthermore, the presence or absence of Mg²⁺ did not affect the RNase digestion profiles, thus indicating that Mg²⁺ did not modify the hammerhead structure significantly to induce self-cleavage. The base-paired stems I and II in the hammerhead structure were **stable** whereas stem III, which was susceptible to digestion, appeared to be an unstable region. The single strand domains separating the stems were susceptible to digestion with the exception of sites adjacent to guanosines; GL2.1 in the stem II loop and G12 in the conserved GAAAC sequence, which separates stems II and III. The absence of digestion at GL2.1 in the stem II **hairpin** loop of the hammerhead complex was maintained in uncomplexed ribozyme and in short oligonucleotides containing only the stem II **hairpin** region. In contrast, the G12 site became susceptible when the ribozyme was not complexed with its substrate. Overall the results are consistent with the role of Mg²⁺ in the hammerhead self-cleavage reaction being catalytic and not structural.

19/3,AB/7 (Item 7 from file: 155)

DIALOG(R) File 155: MEDLINE(R)

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06523321 90148400 PMID: 2856029

Unusual protonated structure in the homopurine.homopyrimidine tract of supercoiled and linearized plasmids recognized by chemical probes.

Vojtiskova M; Palecek E

Institute of Biophysics Czechoslovak Academy of Sciences, Brno.

Journal of biomolecular structure & dynamics (UNITED STATES) Oct 1987, 5 (2) p283-96, ISSN 0739-1102 Journal Code: 8404176

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Plasmid pEJ4, which is a derivative of pUC19 containing an insert with 60-bp-long homopurine.homopyrimidine tract from sea urchin *P. miliaris* histone gene spacer, was studied by chemical probes of the DNA structure osmium tetroxide and glyoxal. The former **probe** reacts with pyrimidine bases, while the latter forms a **stable** product only with guanine residues. These probes can thus be applied as specific probes for the homopyrimidine and homopurine strands, respectively. At pH 6.0 the site-specific modification of the homopurine.homopyrimidine tract by both probes was observed at native superhelical density of the plasmid. In the linear plasmid under the same conditions this modification was absent; it appeared, however, at more acid pH values. In supercoiled DNA the hypersensitivity of the homopurine.homopyrimidine tract to osmium tetroxide

did not substantially change when pH was decreased from 6.0 to 4.0. Changes in NaCl concentration at pH 4.5 did not influence the hypersensitivity to osmium tetroxide; at pH 6.0 this hypersensitivity decreased with increasing NaCl concentration. These results thus show that the chemical probes recognize an unusual protonated structure containing unpaired bases or non-Watson-Crick base pairs. At pH 5.6 the site-specific modification occurred at or near to the middle of the homopurine.homopyrimidine tract, suggesting that a **hairpin** may be involved in the unusual structure under the given conditions. From the models suggested so far for the unusual structure of homopurine.homopyrimidine tracts our results fit best the protonated triplex H form suggest by V.I. Lyamichev, S.M. Mirkin and M.D. Frank-Kamenetskii, J. Biomol. Struct. Dyn. 3,667 (1986).

19/3,AB/8 (Item 8 from file: 155)
DIALOG(R)File 155: MEDLINE(R)
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05965851 88320418 PMID: 3413094
Influence of loop residues on the relative stabilities of DNA
hairpin structures.

Senior M M; Jones R A; Breslauer K J
Department of Chemistry, State University of New Jersey, New Brunswick
08903.

Proceedings of the National Academy of Sciences of the United States of
America (UNITED STATES) Sep 1988, 85 (17) p6242-6, ISSN
0027-8424 Journal Code: 7505876

Contract/Grant No.: GM23509; GM; NIGMS; GM31483; GM; NIGMS; GM34469; GM;
NIGMS

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

We have determined the relative stabilities and melting behaviors of DNA **hairpin** structures as a function of the nonbonded residues in the loop. The specific family of **hairpin** structures we investigated in this work is formed by the 16-mer sequence d[CGAACG(X)4CGTCG], where X is deoxyadenosine, deoxycytidine, deoxyguanosine, or deoxythymidine. As shown below, this 16-mer can fold back on itself to form a family of DNA **hairpin** structures that possess a common hexameric stem duplex and a nonbonded loop of 4 nucleotides. For the **hairpin** structures investigated in this work, we varied the loop composition from all purine residues to all pyrimidine residues. (Formula: see text). We thermodynamically characterized the relative stabilities and melting profiles of these **hairpin** structures by a combination of spectroscopic and calorimetric techniques. To establish a thermodynamic "baseline," we also conducted parallel studies on the isolated hexameric duplex d[CGAACG]..(CG-TTCG)], which corresponds to the common stem duplex present in each **hairpin** structure. Our spectroscopic and calorimetric data reveal the following: (i) The **hairpin** structure with four dT residues in the loop exhibits the highest melting temperature, while the corresponding **hairpin** structure with four dA residues in the loop exhibits the lowest melting temperature. (ii) The free energy data at 25 degrees C reveal the following order of DNA **hairpin** stability for the four structures studied here: T loop greater than C loop greater than G loop greater than A loop. In other words, the pyrimidine-looped **hairpins** of four residues are more stable than the purine-looped **hairpins**. (iii) The loop-dependent order of **hairpin** stability is paralleled by a similar trend in the calorimetrically determined transition enthalpies for **hairpin** disruption. Thus, the enhanced stability of the pyrimidine-looped **hairpin** structures relative to purine-looped **hairpin** structures is enthalpic in origin. To develop insight into the molecular basis for the thermodynamic differences, proton NMR spectroscopy was used to probe

for structural disparities between the most **stable hairpin** structure (T loop) and the least **stable hairpin** structure (A loop). Two-dimensional nuclear Overhauser enhancement spectroscopy revealed connectivities between the residues in the stem duplexes of both **hairpin** structures that are consistent with B-form DNA. In addition, the nonbonded residues in both the T and A loops exhibited the same connectivity patterns. (ABSTRACT TRUNCATED AT 400 WORDS)

19/3,AB/9 (Item 9 from file: 155)

DIALOG(R)File 155: MEDLINE(R)

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04824855 85131184 PMID: 3871776

Two barley alpha-amylase gene families are regulated differently in aleurone cells.

Rogers J C

Journal of biological chemistry (UNITED STATES) Mar 25 1985, 260

(6) p3731-8, ISSN 0021-9258 Journal Code: 2985121R

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Isolation of a full- or nearly full-length cDNA clone for the type B isozyme of barley alpha-amylase is described. The coding region has a high GC content and an unusual codon bias similar to that described for the type A isozyme cDNA previously characterized by our laboratory. The theoretical secondary structure for the 5' end of this type B mRNA, as well as that for the two previously identified type A transcripts, is unusual in that the AUG initiation codon is enclosed in each instance by a very **stable hairpin** loop. Results of primer extension experiments utilizing probes for the 5' coding regions of both types of mRNA are consistent with some of the features of these predicted structures. Primer extension experiments utilizing a **probe** specific for the type B 5' coding region identified only one type of extension products, and these had 5' untranslated region sequences identical to that of the type B cDNA. Thus there appears to be only one major type of type B gene transcribed. Hybridization experiments utilizing 5' probes specific for type A and for the type B mRNA demonstrate different effects of the hormone, gibberellic acid. The type A mRNAs are present in relatively large amounts in unstimulated aleurone cells and increase about 20-fold after stimulation with gibberellic acid. In contrast, the type B mRNA is present at very low levels in unstimulated cells, but increases at least 100-fold after cells are exposed to the hormone.

19/3,AB/10 (Item 1 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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09486896 BIOSIS NO.: 199497495266

Footprinting mRNA-ribosome complexes with chemical probes.

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JOURNAL: EMBO (European Molecular Biology Organization) Journal 13 (16):p 3892-3901 1994

ISSN: 0261-4189

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: We footprinted the interaction of model mRNAs with 30S ribosomal subunits in the presence or absence of tRNA-f-Met or tRNA-Phe using

chemical probes directed at the sugar-phosphate backbone or bases of the mRNAs. When bound to the 30S subunits in the presence of tRNA-f-Met, the sugar-phosphate backbones of gene 32 mRNA and 022 mRNA are protected from hydroxyl radical attack within a region of about 54 nucleotides bounded by positions -35 (+- 2) and +19, extending to position +22 when tRNA-Phe is used. In 70S ribosomes, protection is extended in the 5' direction to about position -39 (+- 2). In the absence of tRNA, the 30S subunit protects only nucleotides -35 (+- 2) to +5. Introduction of a **stable tetraloop hairpin** between positions +10 and +11 of gene 32 mRNA does not interfere with tRNA-f-Met-dependent binding of the mRNA to 30S subunits, but results in loss of protection of the sugar-phosphate backbone of the mRNA downstream of position +5. Using base-specific probes, we find that the Shine-Dalgarno sequence (A-12, A-11, G-10 and G-9) and the initiation codon (A+1, U+2 and G+3) of gene 32 mRNA are strongly protected by 30S subunits in the presence of initiator tRNA. In the presence of tRNA-Phe, the same Shine-Dalgarno bases are protected, as are U+4, U+5 and U+6 of the phenylalanine codon. Interestingly, A-1, immediately preceding the initiation codon, is protected in the complex with 30S subunits and initiator tRNA, while U+2 and G+3 are protected in the complex with tRNA-Phe in the absence of initiator tRNA. Additionally, specific bases upstream from the Shine-Dalgarno region (U-33, G-32 and U-22) as well as 3' to the initiation codon (G+11) are protected by 30S subunits in the presence of either tRNA. These results imply that the mRNA binding site of the 30S subunit covers about 54-57 nucleotides and are consistent with the possibility that the ribosome interacts with mRNA along its sugar-phosphate backbone.

1994

19/3,AB/11 (Item 2 from file: 5)
DIALOG(R) File 5:Biosis Previews(R)
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09069620 BIOSIS NO.: 199497077990
Site-specific cleavage at a DNA bulge by neocarzinostatin chromophore via a novel mechanism.
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JOURNAL: Biochemistry 32 (48):p13138-13145 1993
ISSN: 0006-2960
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The chromophore of the anticancer drug neocarzinostatin (NCS-Chrom) oxidatively cleaves single-stranded or duplex DNA site-specifically in the absence of activating thiol provided that the DNA contains a bulged structure. Point mutations, deletions, and insertions in the DNA analogue and its complement of the 3'-terminus of yeast tRNA-Phe show that for a single-stranded DNA to be cleaved by NCS-Chrom the DNA must generate a **hairpin** structure with an apical loop and at least a two-base-pair stem hinged to a region of duplex structure via a bulge containing a target nucleotide at its 3' side. The size of the loop is not critical so long as it contains at least three nucleotides; the bulge requires a minimum of two nucleotides but must have fewer than five. With a notable exception involving base-pair changes immediately 3' to the bulge, base changes in the bulge and base-pair changes immediately 5' to the bulge retain substrate activity for NCS-Chrom. Maintenance of the bulged structure requires **stable** duplex regions on each side of the bulge. A similar bulged structure, but lacking a loop, formed by the annealing of a linear 8-mer and a 6-mer is

an excellent target for cleavage in the thiol-independent reaction. Drugs such as netropsin, which sequester the DNA into nonbulge containing structures inhibit the reaction. In the absence of O-2 strand cleavage is blocked and quantitatively replaced by a presumed drug-DNA covalent adduct. The cleavage reaction has a pH optimum of about 9 and is much slower than the thiol-dependent reaction. The site-specificity of the cleavage, however, is much greater than in the thiol-dependent reaction. Thus, NCS-Chrom cleaves DNA containing a bulge via a novel mechanism not involving thiol. These studies extend the usefulness of this enediyne as a probe for unusual DNA structures.

1993

19/3, AB/12 (Item 3 from file: 5)
DIALOG(R) File 5:Biosis Previews(R)
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04700505 BIOSIS NO.: 000080003630
2 BARLEY HORDEUM-VULGARE CULTIVAR HIMALAYA ALPHA AMYLASE GENE FAMILIES ARE
REGULATED DIFFERENTLY IN ALEURONE CELLS
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JOURNAL: J BIOL CHEM 260 (6). 1985. 3731-3738. 1985
FULL JOURNAL NAME: Journal of Biological Chemistry
CODEN: JBCHA
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Isolation of a full- or nearly full- length c[complimentary]DNA clone for the type B isozyme of barley .alpha.-amylase is described. The coding region has a high GC content and an unusual codon bias similar to that described for the type A isozyme cDNA previously characterized by this laboratory. The theoretical secondary structure for the 5' end of this type B mRNA, as well as that for the 2 previously identified type A transcripts, is unusual in that the AUG initiation codon is enclosed in each instance by a very **stable hairpin** loop. Results of primer extension experiments utilizing probes for the 5' coding regions of both types of mRNA are consistent with some of the features of these predicted structures. Primer extension experiments utilizing a **probe** specific for the type B 5' coding region identified only 1 type of extension products, and these had 5' untranslated region sequences identical to that of the type B cDNA. Only 1 major type of type B gene is apparently transcribed. Hybridization experiments utilizing 5' probes specific for type A and for the type B mRNA demonstrate different effects of the hormone, GA3. The type A mRNA are present in relatively large amounts in unstimulated aleurone cells and increase .apprx. 20-fold after stimulation with GA3. The type B mRNA is present at very low levels in unstimulated cells, but increases at least 100-fold after cells are exposed to the hormone.

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